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THE USE OF MACROPHYTES FOR HEAVY METAL POLLUTION CONTROL
IN URBAN WETLANDS

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A thesis submitted to the Council for National Academic
Awards in partial fulfilment of the requirements
for the Degree of Master of Philosophy

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The Use of Macrophytes for Heavy Metal Pollution Control in Urban Wetlands

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ABSTRACT

A literature survey has been undertaken to identify the role and function of aquatic macrophytes in the removal of contaminants from polluted surface waters. The survey focussed primarily on heavy metals, however nitrate, phosphate and hydrocarbon removal are also discussed and the application of aquatic macrophytes in wastewater treatment systems is also described.

The objectives of the research are: To determine heavy metal variations in water, sediment and plant tissues in both field and greenhouse conditions and to assess the management implications of macrophytes for heavy metal pollution control in urban wetlands.

The sampling sites and the methods selected for the determination of heavy metal levels in water column, sediment and plant tissues are also discussed.

Heavy metal (Cu, Pb, Zn and Cd) uptake rates and the maximum storage capabilities by sediment and plant have been investigated in controlled greenhouse-based phytoassay experiments. Typha latifolia heavy metal uptake ability is in the order of Cd>Cu>Zn>Pb.

Temporal variations of Cu, Pb, Zn and Cd in urban stormwater runoff, sediment and Typha latifolia tissues at the selected field sites have also been investigated. An increase in sediment and Typha tissue levels is observed from rural to urban sites. Field samples from all sites show a progressive decrease in metal levels from sediment to root to rhizome to leaf. All metals show temporal variability in Typha tissues.

Typha latifolia biomass and plant metal distributions have also been investigated. The rhizome of Typha can store 54 - 61 % and the leaves 32 - 40 % of the total metals found accumulated in the plant. An annual crop of Typha leaf biomass can remove up to Cu 525, Pb 415, Zn 680 and Cd 59 g/ha. Leaf harvesting could provide a useful method of heavy metal removal from recently constructed wetlands.

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Chapter 1: Introduction

1.1 The Research Context

The potential polluting effects of metals in surface runoff have been recognised for a long time (Carpenter, 1924; Jones, 1935). The adverse effects of such discharges on urban aquatic environments have also been documented (Van Hassel et al., 1979; May and Mckinney, 1981). Metal contamination of receiving waters has been identified as the cause of both fisheries and faunal depletion (Mance, 1987) and also can present hazards to public health (Kobayshi, 1971).

The most frequently adopted 'single-shot' solution to the urban runoff problem is to provide on-site storage facilities which potentially can offer the dual purpose function of both quantity and quality control (Hall et al, 1990). Whilst the large and highly variable volumes presented to such storage basins will generally render unfeasible any application of the sort of treatment processes used for sanitary or industrial effluents, the combined possibilities of detention and biofiltration contact treatment provide considerable least-cost opportunities for urban stormwater management and pollution control. The experience of root zone biotechnology (RZB) systems may provide the unique advantage of enabling primary filtration and secondary biological uptake for both solid and soluble pollutant phases through the judicious use of macrophytic planting programmes (Cooper et al., 1989). RZB systems are discussed further in Chapter 2 where their role as pollution scavengers is developed in further detail.

Even though COD, BOD and nutrient removal by RZB systems have been extensively studied, the role of macrophytes in heavy metal uptake and removal has not been fully investigated. The study of the efficiency and pathways whereby metal removal by macrophytes occurs in stormwater runoff might therefore potentially provide a useful method for heavy metal pollution control in urban wetlands. It is within this general pollution context that the current research project has been specifically devised and developed.

The research has been carried out with the following major objectives in mind:

- a) To determine heavy metal variations in urban stormwater runoff, sediment and plant tissues across selected field sites and to establish relationships between these 'carrier' phases.
- b) To determine heavy metal uptake efficiency and maximum storage capabilities of plants under controlled greenhouse-based phytoassay experimental conditions.
- c) To assess the management implications of macrophytes for heavy metal pollution control in urban wetlands.

1.2 The Metals Selected for Investigation

The concentrations and loadings of copper (Cu), lead (Pb), zinc (Zn) and cadmium (Cd) in urban stormwater runoff, sediment (or peat in the case of greenhouse experiments), plant root, rhizome and leaf tissues were investigated during this research project. It was impossible to analyse a further range of heavy metals in the time required and

therefore this study has been focused on the above-mentioned metals.

Cu, Pb, Zn and Cd comprise those metals which present the greatest overall hazard to the aquatic environment especially to fish and vertebrates (Dryssen et al., 1971, Mance, 1987). Lead is a widely used metal and a major vehicle derived pollutant which is a neurotoxin and has given rise to much public concern (Jaworski, 1979; USEPA, 1980). It is associated in particular with the urban environment (WHO, 1989). Cadmium is a highly toxic metal which can cause kidney disease in humans and this has stimulated considerable scientific attention (Kobayshi, 1971; Friberg et al., 1972). Copper and Zinc are also widely used metals in the urban industrial environment and are therefore commonly encountered pollutants in urbanised catchments (Mance, 1987). Although both metals are essential elements for plant, animal and human nutrition, they are toxic at certain threshold levels to many aquatic organisms.

1.3 The Plants Selected for Investigation

Typha latifolia, an aquatic macrophyte, was selected as the main species for the study project with Juncus effusus also being used for greenhouse experiments. Typha latifolia was selected to monitor metal uptake in preference to Phragmites sp on account of its wide distribution and abundance in urban wetlands and its ability to survive transplanting to laboratory conditions.

Typha latifolia is a native emergent species which grows in shallow water, especially on inorganic substrates or where there is silting and rapid decay of organic matter,

in lakes, ponds, canals and slow-flowing rivers. It is widely distributed in suitable habitats throughout Britain, though it occurs less frequently in the north. Typha species colonize new sites by seed dispersal, with seedling establishment being largely restricted to areas of exposed wet mud (Valk and Davis, 1976; Grace, 1984). Seeds are very small, disperse great distances by wind, and are produced in abundance with values of 222,000 seeds per inflorescence reported (Yeo, 1964). Typha latifolia is a good pre-emptive competitor when the plants are larger and at low densities (Grace, 1987), and has a great capacity for vegetative spread (McNaughton, 1975) and a great proportion of its biomass occurs below ground. One plant of Typha can spread by its rhizome over areas as great as 60 m² within 2 years of germination (Dykyjova and Kvet, 1978). After a habitat is fully colonized, seed germination may be inhibited (Grace, 1984; Grace and Harrison, 1986), and the stand is maintained by vegetative reproduction. In some marshes the Typha is not rooted in the mineral soil but floats on a fibrous mat. It is thought that buoyancy of the mat is mainly due to the Typha rhizome having a lower specific gravity than water and to gases that are trapped in the floating mat. The habitat conditions on this floating mat appear to change little over long periods and in time Typha may eventually form almost pure stands, expand, and coalesce to form a complete cover (Krusi and Wein, 1988). Typha latifolia plants have a great ability to survive transplanting. Studies showed that after transplanting by means of rhizome and shoot clumps, the growth of the plants is excellent (Kyte, 1984).

Juncus effusus is a British native soft rush. It is very abundant and locally dominant in wet pastures, bogs and

damp woods, especially on acid soils. This plant was selected for this research study because of the persistence of its foliage during the winter and its ability to survive transplanting.

1.4 Thesis Content and Organisation

The structure of the thesis and a fuller description of the research programme is outlined below. Following this introductory section, Chapter 2 provides a review of the literature which gives further and more detailed background information for the research project. The major sources and the toxicity of heavy metals found in urban wetlands along with the mechanisms of plant heavy metal uptake, removal and release are described. The use of macrophytes for nutrient and hydrocarbon removal and the application of aquatic macrophytes as wastewater treatment systems are also briefly discussed. Chapter 3 provides the details of sampling locations, sampling methods and analytical techniques which have been employed in the research.

Chapters 4 and 5 detail the two main aspects of the experimental work undertaken in the course of the research. Chapter 4 describes the greenhouse dosing experiments which investigate and analyse both peat and plant metal uptake efficiency and maximum storage capabilities. Chapter 5 describes the investigation undertaken to determine heavy metal levels in urban stormwater runoff, sediment and plant tissues under field conditions and the relationships established between them. Chapter 6 compares the results of greenhouse dosing experiments and field studies and develops the management implications of macrophytes for heavy metal pollution

control. The discussions presented in Chapter 4, 5 and 6 also form part of two co-authored publications entitled "Metal Uptake and Associated Pollution Control by Typha latifolia in Urban Wetlands" (Zhang et al, 1990), and "The Use of Macrophytes for Water Pollution Control in China and UK" (Zhang et al, 1990). These articles are included as appendices to the thesis. The concluding Chapter 7 provides a general overview of the main conclusions and suggestions for further work.

Chapter 2: A Review of the Use of Aquatic Macrophytes in Water Pollution Control

2.1 Introduction

Aquatic macrophytes have been extensively studied over the last few decades in terms of their ability to take up and absorb pollutants, and therefore indirectly provide an indication of water quality (Haslam, 1982; Wang, 1986). It has been found that many species of aquatic macrophytes can grow rapidly and exhibit a tolerance of polluted water (Haslam and Wolseley, 1981). They can take up a variety of pollutants quickly and can be easily harvested, thereby providing both a temporal measure of pollution and a method of removing pollutants from the aquatic system. A number of studies have been carried out during the last 15 years defining the use of aquatic macrophytes in water pollution control (Mortimer, 1985; Reddy and DeBusk, 1987). In particular, recent research has been directed toward the use of aquatic macrophytes for wastewater treatment (Middlebrooks and Reed, 1981; Arthur, 1986; Cooper et al., 1989).

This review focuses on the ability of particular species of aquatic macrophytes to remove contaminants from polluted surface waters. The principle focus is on heavy metals, but other pollutants such as nitrate, phosphate and hydrocarbon removal are briefly discussed and the application of aquatic macrophytes as wastewater treatment systems is also described.

2.2. Heavy Metal Removal

2.2.1 Sources of Heavy Metals

The sources of heavy metals found within urban wetlands are both natural and anthropogenic. Natural sources include weathering of rocks, impervious surfaces and soils. The anthropomorphic influences include industrial activities, such as smelting, metal working; mining and metal extraction processes (Lagerwerff and Brower, 1975; Davies and Ginnever, 1979); domestic sources, such as paint, house dust, printed matter, paper, textiles and plastics (Sturges and Harrison, 1985) and vehicle exhausts which are particularly evident in the urban environment (Harrison, 1979).

The contributing sources of heavy metals to wetlands include suspended river loads; storm runoff; sewer discharges and atmospheric deposition (Ellis, 1976; Eaton, 1979; Revitt and Ellis, 1980). Street dusts often contain elevated concentrations of a range of toxic metals (Sartor and Boyd, 1972) which can be discharged from surface water sewers to urban wetland environments. In some instances these dusts represent a significant pollutant source, especially when stormwater runoff removes the street materials and their associated metal from the roadway causing an increased metal input to rivers (Harrison et al., 1981; Ellis and Harrop, 1984).

2.2.2 Heavy Metals in the Aquatic Environment

The flows of energy and matter in an aquatic ecosystem are shown in Fig. 2.1. In such aquatic systems, heavy metals are partitioned between different compartments of the ecosystem such as water, sediment and biota (Moore and Ramamoorthy, 1984; Prahalad and Seenayya, 1989). In water, which is the primary contaminated medium, a high degree of variation in metal concentration arises (Forstner and Wittman, 1979). Metals that do not remain soluble in water are sorbed and accumulated by bottom sediments acting as a sink (Hakanson, 1980). Sediments play a crucial role in water quality. Although they may remove pollutants from the water column, pollutants which have accumulated in the sediments may provide the surface water (long after the source of pollution has ceased) with contaminants (Salomons, 1985). The content of heavy metals in contaminated sediment is normally much higher than in water and plants (Ellis, 1986). The amount of heavy metals present in the sediments may thus be considered to be the amount potentially available to the plants.

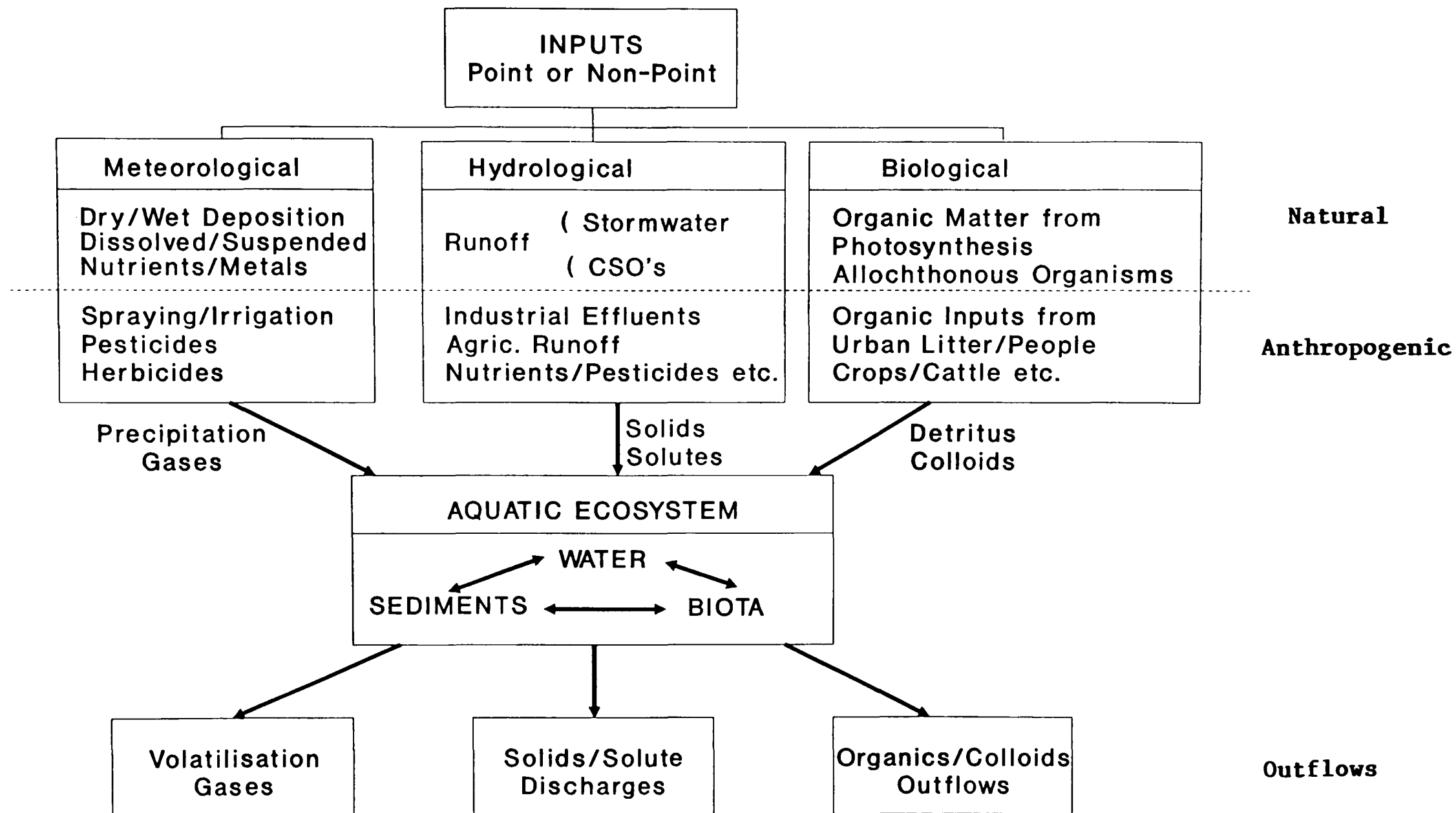


Fig.2.1 The Flows of Energy and Matter in Aquatic Ecosystems

2.2.3 Heavy Metal Speciation

The environmental mobility and bioavailability of a metal is dependent upon the physico-chemical forms in which the metal is associated with sediment and water. The classifications of metals associated with dusts, sediments and soils are given in Table 2.1. A physico-chemical speciation scheme has been applied to the analysis of Cu, Pb, Zn and Cd in urban stormwater (Morrison et al., 1984). According to this study, Zn and Cd exhibit a preference for the dissolved phase whereas Pb predominates in the suspended solid phase. Copper tends to be distributed equally between both phases. The potentially toxic forms of the metals, (the bioavailable metals) in the dissolved phase (electrochemically available) and in the exchangeable particulate phase are said to account for about 63% of the total Zn, 77% of the total Cd, 66% of the total Pb and 32% of the total Cu.

In overall terms the results of Harrison et al.'s study (1981) of chemical associations in street dusts and roadside soils suggest that the mobility and bioavailability of the four metals probably declines in the following order: Cd>Pb Zn>Cu, whereas Ellis et al., (1986) suggests that the overall availability of street surface sediment associated metals to the dissolved phase of runoff is Cd>Zn, Pb>Cu.

Table 2.1: Classification of Metals Associated with
Dusts, Sediment, and Soils (from Harrison et al., 1981)

classification	form of association*	extraction technique
soluble	metal ppt; pore water	release to pure water or river water
exchangeable	specifically adsorbed, ion exchangeable	exchange with excess cations
carbonate phase	ppt or co-ppt	release by mild acid
Fe-Mn oxide phase	specifically adsorbed, co-ppt	reduction
organic phase	complexed; adsorbed	oxidation
residual	in mineral lattices	digestion with strong acid
*ppt = precipitate.		

2.2.4 The Toxicity of Heavy Metals

The concentrations of all elements in soil will be reflected to some extent in the dry matter of plants which grow in them (Baker and Walker, 1989). High levels of heavy metals in sediment and plant can be toxic. The background and toxic metal concentrations of soil and plants are given in Table 2.2. It has been found that soil pollution by heavy metals can restrict the growth of plants. A reduction of biomass production and nutritional quality is observed in crops grown in soil contaminated

with moderate levels of heavy metals (Cottenie et al., 1976; Lepp, 1981). These elements generally inhibit physiological processes, e.g. photosynthesis (Carlson et al., 1975; Clijsters and Van Assche, 1985), phloem translocation and transpiration (Carlson et al., 1975); respiration is less sensitive (Lee et al., 1977b; Van Assche et al., 1979).

Zinc. Zn has long been known as an essential element in plant nutrition. It acts either as a metal component of enzymes (such as alcohol dehydrogenase, superoxide dismutase, carbonic anhydrase and RNA polymerase) or as a functional, structural or regulatory co-factor of a large number of enzymes (Price et al., 1972; Jyung, 1975; Marschner, 1986).

Table 2.2: The Background and the Toxic Levels of Heavy Metals
(from Romero et al., 1989; Baker and Brooks, 1989)

Soils (mg/kg)			Plants (mg/kg)	
	Unpolluted	toxic level	Unpolluted	toxic level
Pb	25	100	10	150
Cd	1	5	-	5
Cu	25	100	12	40
Zn	65	300	60	400

High Zn levels in soil (or sediment) can be toxic to the plants. Zn toxicity results in a reduction in root and shoot growth (Van Assche et al., 1988, Allus et al., 1988) and leaf expansion which is followed by chlorosis. High levels of Zn in the nutrient medium depress the uptake of P and Fe (Adriano et al., 1971).

Copper. Cu is also an essential plant micronutrient being a component of the protein structure of a range of enzymes involved in electron transport and redox reactions in mitochondria, chloroplasts, cell walls and the cytoplasm of plant cells. Other Cu proteins play important roles in carbohydrate and nitrogen metabolism, and lignification of the cell wall (Boardman, 1975; Marschner, 1986).

For most plant species high amounts of Cu in the nutrient medium are toxic to growth. The effect appears to relate in part to the ability of Cu to displace other metal ions and particularly Fe from physiologically important centres. Chlorosis is thus a commonly observed symptom of Cu toxicity (Daniels et al., 1972). The inhibition of root growth is one of the most rapid responses to toxic Cu levels (Wainwright and Woolhouse, 1975).

Lead. Lead is neither an essential nor beneficial element in plant nutrition. Pb is a major chemical pollutant of the environment and is highly toxic to man. The reason for Pb toxicity in plants is not clear. However, the organic form trialkyl Pb is a powerful mutagenic agent and is known to derange the spindle fibre mechanism of cell division in plant cells (Bryce-Smith, 1975).

Cadmium. Cd is toxic to plants. The basic cause of the toxicity probably lies in the much higher affinity of Cd for thiol groupings in enzymes and other proteins. The presence of Cd therefore disturbs enzyme activity (Mengel and Kirkby, 1978).

2.2.5 Heavy Metal Uptake by Macrophytes

Most macrophyte species can take up heavy metals and some of them can accumulate heavy metals to high levels (Adams et al., 1973; Tokunaga et al., 1976; Chigbo et al., 1982; Sridhar, 1986; Lan et al., 1990). Chigbo et al. (1982) found that the exposure of the water hyacinth (Eichhornia crassipes) to Cd levels of 10 mg/l for 2 days resulted in a stem concentration of 501 mg/kg and a leaf concentration of 574 mg/kg. Muramoto and Oki (1983) reported that after exposure to a 8.0 mg/l Pb solution for 16 days, the Pb levels in water hyacinth roots reached 25790 mg/kg and 1810 mg/kg in the tops of the plant. The maximum heavy metal removal values by per individual plant are shown in Table 2.3.

Cu bioaccumulation in water lilies (Nuphar lutea) has been shown to reach levels of up to 110 mg/kg in petioles according to a field study carried out by Aulio (1980). Blake et al. (1987) reported that after one month of contact with sediment (initial concentration of Zn in the medium was 10 mg/kg), Zn levels in Typha latifolia roots reached up to 1400 mg/kg. After contact with culture medium (without sediment) for one month (initial concentration of Zn was 1-10 mg/l), the Zn levels in Typha latifolia roots reached up to 3600 mg/kg (Table 2.4). Pinto et al. (1987) showed that after cultivation of water hyacinth plants for 24 hours in a 40 mg/l silver

solution, the average concentration of silver recovered from the dried plant material was 70% of the initial silver concentration of the solution with a purity of 98%. This high uptake implies extremely high tolerance to specific elements as confirmed by Baker and Brooks (1989) in their study of the hyper-accumulation of metallic elements by plants.

These investigations would therefore imply a basic ability of macrophytes for indicating anthropogenic pollution and also in removing pollutants from wastewater.

Table 2.3: Maximum heavy metal removal values by water hyacinth (from Muramoto, 1983)

treatment	Maximum content per individual plants (g/kg)	Mean values (g/m ²)	Maximum values (g/m ²)
Cd 1.0 ppm	0.146	1.46	6.56
4.0	0.393	3.93	17.7
8.0	0.124	1.24	5.60
Pb 1.0 ppm	1.05	10.5	47.5
4.0	7.46	74.6	336
8.0	13.9	139	627
Hg 0.5 ppm	0.007	0.77	3.47
1.0	2.34	23.4	105
2.0	0.17	1.70	7.52

Table 2.4: Comparative increases of concentrations of Zn in Typha latifolia (from Blake et al., 1987)

Zn Compound	Concentration	Main Plant		New Plant		
		Roots	Rhizome	Roots	Rhizome	Leaves
mg/kg of tissue (dry wt)						
Zn Chloride	1 mg/l (A)	650	100	2500	390	68
	10 mg/l (B)	2400	270	3600	1800	250
	Ratio (B)/(A)	3.7	2.7	1.4	4.6	3.7
Zn-EDTA	1 mg/l (C)	310	26	380	200	30
	10 mg/l (D)	1000	80	1500	1100	350
	Ratio (D)/(C)	3.2	3.1	3.9	5.5	11.7

Harrison and Chirgawi's research (1989) of both soil and air as contributors of some trace metals to plants further suggests that the efficiency of soil uptake and atmospheric deposition are in the order of Zn>Cd>Ni>Cr>Pb (sometimes Cd>Zn), while Xian's (1989) study of the response of the kidney bean to concentration and chemical form of Cd, Zn and Pb in polluted soil shows that the uptake of metals was according to their solubility Cd>Zn>Pb. This latter hierarchy confirms the earlier work of Ellis et al., (1986) referred to in section 2.2.3.

2.2.6 The Effects of Heavy Metal Uptake on Macrophytes

Variation in the long-term metal uptake rate has been found to depend upon the species of plant, with higher heavy metal contents being associated with submerged species such as Elodea sp. and Callitriche sp. compared to floating species such as Lemna sp. and Spirodela sp.

(Werff and Pruyt, 1982) This suggests that high biomass species rooted in the sediment have a greater ability to remove pollutants than species exposed to the water column alone. Some existing studies (Merchyulenene and Nyanishkene, 1976; Leland and McNurny, 1984) strongly suggest that submerged macrophytes are capable of attaining tissue heavy metal concentrations far in excess of concentrations in the water column.

Plants appear to accumulate a higher concentration of metals in the roots than in the stems and leaves (Reimer, 1989). Welsh and Denny (1980) as well as Heisey and Damman (1982) have reported root to shoot ratios for lead concentrations of 3.8 - 5.8 and 3:1, respectively.

Uptake rates are also dependent upon changes in the seasonal growth rate. Actively growing plants of any species absorb metallic ions much faster than old or dying or seasonally decomposing parts of plants. For example, Zn levels in the leaves and stems of Phragmites australis and Cu concentrations in the stems are at a maximum during the growing season and decrease thereafter (Larsen and Schierup, 1981). Concentrations of Pb in the leaves have also been found to increase during and after the growing season (Fig. 2.2). The uptake rate is also dependent upon the specific metal ion being absorbed (Mortimer, 1985). The efficiency of uptake of metals from soil is generally thought to be high for Zn and Cd and low for Pb (Harrison and Chirgawi, 1989).

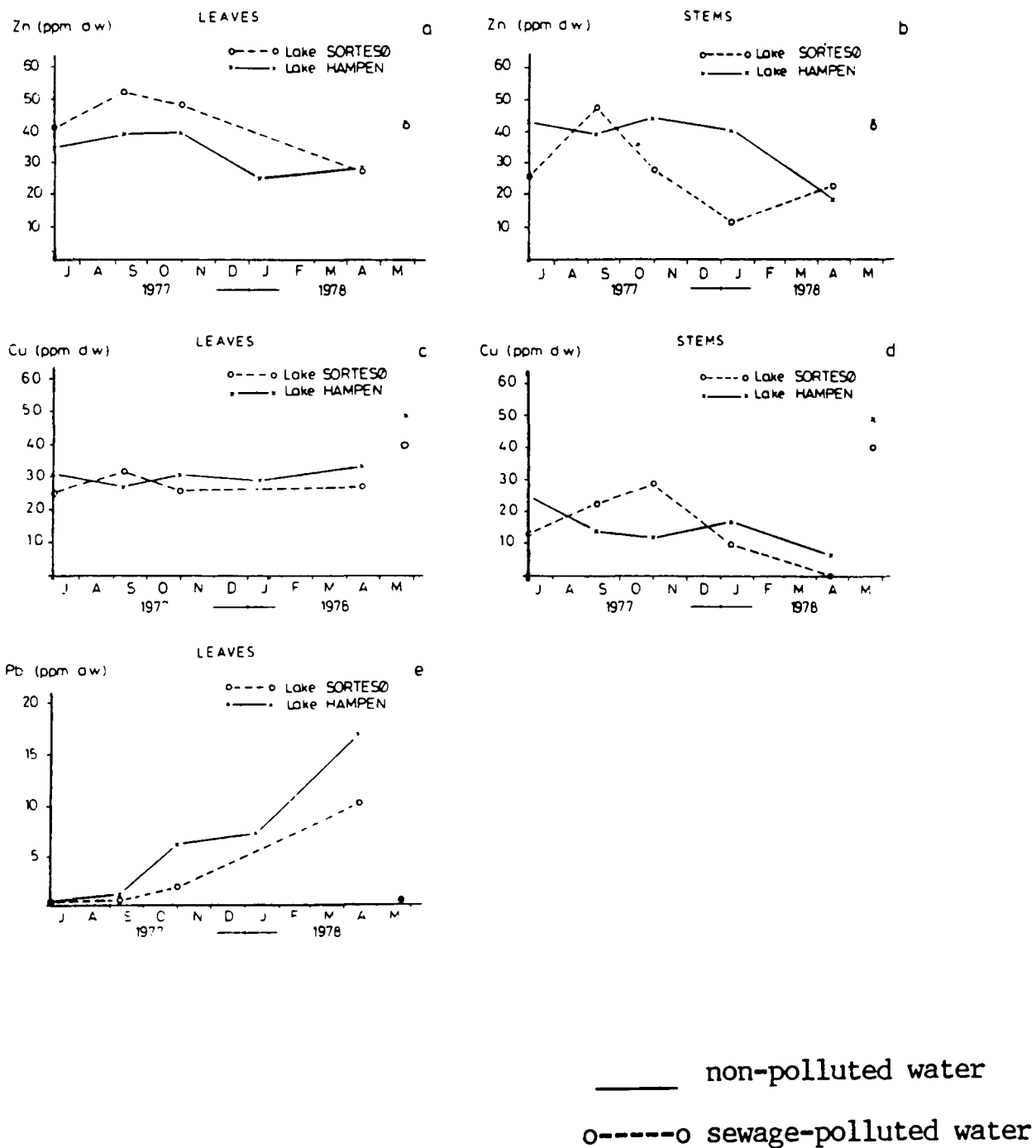


Fig.2.2 Seasonal variation in concentrations of Zn, Cu and Pb in leaves (a,c,e) and stems (b,d) of Phragmites australis (from Larsen and Schierup, 1981).

There are some fairly obvious factors which are known to affect the efficiency of metal uptake including root mass, pH, chelating agent concentration and the presence of other competing metals. Hardy and Raber's study (1985) of Cd uptake by water hyacinth demonstrate that plants with greater root mass remove Cd more efficiently from solution per unit time. This study also shows that there is a sharp increase in the initial rapid uptake at a pH of about 5. Nitrilotriacetic (NTA), N - (2 - hydroxyethyl) ethylenedinitrilo - N, N', N' - triacetic acid (HEDTA), ethylenedinitrilo - N, N, N', N' - tetraacetic acid (EDTA) and trans - 1, 2 - cyclohexylenedinitrilotetraacetic acid (CDTA) significantly reduced the rate of Cd and Zn uptake by water hyacinth (Hardy, 1985). There are six metals which extensively hindered uptake of the Zn. These are Fe^{3+} , Cu^{2+} , Hg^{2+} , Co^{2+} , Cd^{2+} and Ni^{2+} .

Since ammonium ions, metal ions, chelating agents and pH levels can vary considerably in polluted water, the condition of the water with reference to these parameters must also be taken into consideration when macrophytes are to be used as an indicator of water pollution or to purify wastewater (Nasu and Kngimoto, 1983; Hardy and Raber, 1985).

2.2.7 The Mechanisms of Heavy Metals Uptake

River plants take up metals from both sediment and water, sediment usually being the more important source (Haslam, 1987). Emergent plants on the bank eg. Phragmites communis obtain all their minerals from the sediment. They absorb little minerals through their thick rhizome or aerial parts. So effective mineral uptake occurs only when living roots are present (Haslam, 1987).

Heavy metals can be taken up either from a sediment (soil)-root pathway or foliar route. On the one hand, heavy metals are taken up from sediments by roots and translocated to the rhizomes and aerial parts of emergent species. The greater part of heavy metals taken up by the plants would appear to be retained in the roots and rhizomes, with only small amounts being translocated to the above-ground part of the plant. For instance, up to 77 to 97% of heavy metals taken up by Phragmites communis is retained in the roots and rhizomes according to Larsen and Schierup (1981) and about 80% of the total cadmium uptake by water hyacinth after 30 days exposure was found in the roots by Blake et al. (1987).

However, analysis of the metals in the plant tissues (Harrison and Chirgawi, 1989) showed that the foliar route is potentially of similar importance to the soil-root pathway as a route of transport to the exposed parts of the plants. Metals entering the plant from the atmosphere can be translocated to unexposed portions of some plants, eg. radish and turnip. The exposed parts showed the highest metal accumulation with lower values found for unexposed plant parts, indicative of translocation of atmospherically-derived metal within the plant by mechanisms similar to those for soil-derived metal. Experiments with rainwater show a generally positive wet deposition component, rather than the contribution of dry deposition to the metal content of the plant.

Plants can grow on toxic sediment or soil: either they avoid their uptake and restrict the transport of the metal, or they have an internal mechanism that renders the metal harmless (Baker and Walker, 1990). Tolerant plants have high concentrations of metals in their tissues, eg.

Eichhornia crassipes (Chigbo et al., 1982) and Typha latifolia (Blake et al., 1987). Clearly, they must have evolved a way to protect their life processes. Plant physiologists have proposed a number of mechanisms (Farago et al., 1975; Kelly et al., 1975; Kersten et al., 1980; Baker and Walker, 1990). One suggestion is that once the metal enters plant cells, compounds that bind to the metal in the cytoplasm are produced. Once bound, the metal complex may stay in the cytoplasm or move to another part of the cell, preventing further contact with metabolic processes. In either case, the toxicity of the metal is greatly reduced. Compounds suggested for the binding role in the vacuole include organic acids and sugar phosphates; in the cytoplasm the probable binders are phytochelatins (Thurman and Hardwick, 1988).

Zinc taken up by plants from soil and sediment is translocated in either free ionic form or as simple organic acid complexes to the shoots. Accumulation of Pb by the foliage of plants is an exceedingly rare event due to the readiness with which this element can be precipitated as the insoluble sulphate in the rhizosphere, hence minimising potential uptake and transport to the aerial parts of the plants (Baker and Brooks, 1989).

2.2.8 Heavy Metal Removal

The partial removal of heavy metal pollutants by macrophytes can be achieved by frequent harvesting of the foliage and stems where a proportion of the accumulated metals are stored in the plant biomass. The potential rate of pollutant storage by an aquatic plant is limited by the growth rate and plant biomass per unit area as well as by uptake rate. The ease of harvesting macrophytes may be of

some significance in terms of their use as a wastewater purification technique, though this is probably less important than the capacity of the whole system to act as a sink. Muramoto and Oki (1983) reported that the maximum values of metal removed by water hyacinth are: 177 kg/ha from water containing 4.0 mg/l Cd, 6,270 kg/ha from water containing 8.0 mg/l Pb, and 1,050 kg/ha from water containing 1.0 mg/l Hg (Table 2.3).

If plants are not harvested, heavy metals can be released to the surrounding aquatic environment by plant decomposition. Black et al.'s studies (1979, 1984) showed that 25% to 42% Zn accumulated in Typha latifolia leaves and potentially could be released from the decomposed leaves. It is therefore necessary to cut and harvest the plant, as this technique allows the export of 25 % to 42 % of the Zn accumulated in Typha latifolia and prevents the leaching of the metal from decaying organic matter. DeBusk and Reddy's study (1990) shows that after injecting Pb (900 mg with concentration of 1.0 mg/l) and Cu (2250 mg with concentration of 2.5 mg/l) continuously for two months into raceways containing pennywort (Hydrocotyle umbellata), approximately 69% of the 14.0 g/m² Cu and 85% of the 4.9 g/m² Pb applied were removed with effluent concentrations averages 839 ug Cu/l and 149 ug Pb/l.

2.2.8 Heavy Metal Release

In order to understand the heavy metal cycle in an aquatic environment, it is very important to have a knowledge of the decomposition process. Several reports have been published on the behaviour of heavy metals in the decomposing litter contained within aquatic environments. Larsen and Schierup (1981) found that heavy metals tend to

show an increase in concentration in decomposing Phragmites sp. litter during decomposition. From a one year study in a non-polluted lake, concentrations of Zn, Cu, Pb and Cd (mg/kg oven dry weight) in Phragmites leaves increased from 108.8, 5.0, 5.5 and <0.05 to 135.2, 17.0, 13.6 and 0.56, respectively.

By comparing the concentration of the different heavy metals in litter during decomposition to the concentration of the plant tissues prior to decomposition, heavy metals were observed to behave in different ways. The observed increase in metal concentration may arise as a result of a greater decrease in biomass compared to metal weight during decomposition and therefore does not necessarily relate to an actual increase in total metal load. During the decomposition of Phragmites sp. the relative contents of Zn and Cu in decomposing litter were either similar to that prior to decomposition or significantly lower. This might be a consequence of the leaching of Zn and Cu being easier under the specific aquatic conditions being studied than for Cd and Pb. The relative contents of Pb and Cd in partly decomposed Phragmites sp. leaves were however, found to be significantly above those prior to decomposition. The accumulated Pb was presumably absorbed from the surrounding water column or derived from a concentration of the metal following a reduction in biomass.

2.2.10 Biological Accumulation of Heavy Metals in the Marine Environment

Industrial and demographic growth along coastlines poses a threat, principally to estuaries, which are primary recipients of pollutants and critical intermediates in the

biological cycle of marine organisms (Seeliger and Lacerda, 1986). The biological accumulation of heavy metals in marine environments has become a widely accepted concept. Heavy metal concentrations in benthic marine algae are magnified several thousand times with respect to their concentrations in seawater (Table 2.5). Due to their pollution tolerance, macroalgae have been considered the most suitable group of organisms for biological metal monitoring in coastal waters (Seegier and Cordazzo, 1982).

Table 2.5: Concentration factors ($\times 10^4$) in benthic algae tissue (ug/g dry wt) and dissolved metals in water (ug/ml)

Metal	Algal genera				Source
	Blidingia	Enteromorpha	Ulva	Fucus	
Cu	0.71-1.83	0.56-0.77	0.47-0.86 0.12	0.36-0.74	Seeliger(1977)
					Yamamoto(1972)
				0.25	Black(1952)
				0.45	Preston(1972)
Pb	2.7-8.2	2.0-4.5	1.7-4.9	0.45-2.7	Bryan(1973)
				1.3-2.4	Seeliger(1977)
				0.087	Black(1952)
				0.23	Preston(1972)
				0.24	Bryan(1973)

Metal accumulation by algae is, at least in its first stage, a passive physico-chemical procedure occurring on the surface of algal cells. The procedure involves the binding of metal ions within the cell wall matrix, which contains anionic groups, with large cation exchange capacity (Reed and Darrig, 1983). The large biomass of

benthic algae in coastal waters will eventually undergo decomposition and ultimately be mineralized (Riley , 1970). Traditionally, in studies of biological magnification the primary producers have been considered to be consumed directly by herbivores, and thus their metal complement passed directly into the next link in the food web. However, in most coastal waters the benthic algae are largely unconsumed and their organic matter is passed along the food web as detritus and dissolved organic matter (Seeliger, 1979). It has been suggested that heavy metal concentrations in seaweeds are not subject to short term erratic fluctuations as are metals in water, but integrate the metal concentrations in water over relatively long intervals of time. This makes them valuable tools for the monitoring of heavy metal levels in the ambient water over extended periods of time (Seeliger, 1979).

2.3 Nutrient Pollution Removal

Nutrient pollution has been the source of a number of environmental problems, even though dissolved mineral nutrients such as calcium, nitrogen, phosphorus and potassium are essential for the maintenance of the production of aquatic plants and animals. The overload of nutrients to receiving waters has caused a rise in the primary plant productivity which has resulted in a reduction of the oxygen in the water and the death of fish. The minerals are taken up from both the water and soil and accumulate in roots, stems and leaves (Haslam, 1987). As a result macrophytes participate actively in the biological removal of nutrients, retaining and accumulating them in aquatic environments. It is now firmly established that macrophytes remove many biogenic

elements and act as accumulators of nutrients (Kovalskii and Grivovskaya, 1976; Sundblag and Wittgren, 1989).

2.3.1 The Effects of Nutrient Uptake

Tsutomu and Teranish (1988) showed that the total N uptake rate was correlated with the N concentration by a Michaelis-Menten type equation, in which the maximum uptake rate of N was expressed as a function of the temperature and plant density. The P uptake rate however was influenced by not only the P concentration, temperature and plant density, but also by variation in the P content of the plant. The accumulating ability of macrophytes shows species variation and depends on the growth location and season (Morozov, 1984). The removal of biogenic substances from water by macrophytes, which is always at a maximum rate at the start of the growing season, decreases by the end of the growing season as a result of losses due to excretion from plant tissues back to the environment. For example, the common reed Phragmites communis with a dry mass of up to 3 kg/m² during growth, can extract about 45g nitrogen and 18g phosphorus from water per annum. Narrow-leaved reedmace Typha angustifolia with a dry mass of 3.6 kg/m² can extract 32 g phosphorus per annum (Orekhovskii, 1986). Laboratory experiments with wastewater have shown that narrow-leaved reedmace and the water-weed, Elodea canadensis, can extract up to 60% of the ammoniacal nitrogen, more than 80% of the nitrates, up to 99.98% of nitrites and 37 - 57% of the phosphorus over a period of 1 week (Morozov, 1984). Water hyacinth Eichhornia crassipes can remove 71.3% ammoniacal nitrogen, 50% phosphorus and 30% potassium after 5 days (Xie et al., 1984).

2.3.2 Nutrient Storage and Release

Studies have shown that more than 50% of nutrients extracted by macrophytes were stored in below ground portions of the plants (Reddy and DeBusk, 1987). Emergent macrophytes have more supportive tissues than floating ones and so they have greater potential for storing the nutrients over a longer period. Typha latifolia can absorb 600 - 2630 kg/ha/yr of nitrogen and 75 - 403 kg/ha/yr of phosphorus. In addition, 250 - 1560 kg/ha and 45 - 375 kg/ha nitrogen and phosphorus, respectively, can be stored (Table 2.6).

Table 2.6: Storage of nitrogen and phosphorus and rate of plant uptake for selected aquatic macrophytes
(from Reddy and DeBusk, 1987)

Plant	Nitrogen		Phosphorus	
	Storage	Uptake	Storage	Uptake
	kg/ha	kg/ha/yr	kg/ha	kg/ha/yr
FLOATING MACROPHYTES:				
<u>Eichhornia</u>				
<u>crassipes</u>	300-900	1950-5850	60-180	350-1125
<u>Pistia stratiotes</u>	90-250	1350-5110	20-57	300-1100
<u>Hydrocotyle</u>				
<u>umbellata</u>	90-300	540-3200	23-75	130-770
<u>Alternanthera</u>				
<u>philoxeroides</u>	240-425	1400-4500	30-53	175-570
<u>Lemna minor</u>	4-50	350-1200	1-16	116-400
<u>Salvinia rotundifolia</u>	15-90	350-1700	4-24	92-450
EMERGENT MACROPHYTES:				
<u>Typha</u> spp.	250-1560	600-2630	45-375	75-403
<u>Juncus</u>	200-300	800	40	110
<u>Scirpus</u>	175-530	125	40-110	18
<u>Phragmites</u>	140-430	225	14-53	35

Plant species which have a higher relative biomass can store higher nutrient load. Thus, in order to improve water quality, the most efficient way to remove nutrients is to remove the biomass by harvesting the plants (Frederiksen, 1987). If plants are not harvested, the dead tissue will decompose rapidly and release nutrients into the water. Frequent harvesting of biomass is therefore essential and necessary to avoid losses of nutrients back to the water column. Macrophytes essentially extract all of the nutrients they require for their growth entirely from the benthic sediment and release the nutrients during decomposition back to the water. So the life cycle of macrophytes may provide an important source of nutrients to the aquatic environment. When a high biomass exists, macrophytes can play an important part in nutrient cycles of aquatic ecosystems (Jewell, 1971; Howard-Williams and Davies, 1979; Carpenter 1980, 1981; Werner and Adams, 1984).

2.4 Hydrocarbon Pollutant Removal

2.4.1 Hydrocarbon Degradation

Hydrocarbon degradation is achieved through bacterial activity. Oxygen is therefore a very important factor as it affects microbial growth. Aquatic plants have the unique feature of transporting O_2 through the leaves, stems and roots. Transported oxygen enters sediment interstitial water, and is utilised by aerobic bacteria during the oxidation of hydrocarbons (Foght and Westlake, 1987). Oxygen transferred by plants into the root zone therefore plays a significant role in supporting the aerobic bacteria in this area and in the degradation

of hydrocarbons. For example, 0.02 - 0.1 g of the root mass of Typha latifolia can transport 1.39 ± 1.49 mg O_2 /g/hr and 0.03 - 0.06 g of the root mass of Sagittaria latifolia can transport 1.72 ± 0.87 mg O_2 /g/hr (Table 2.7).

Table 2.7: Oxygen transport through selected aquatic macrophytes (from Moorhead and Reddy, 1987)

Plant	Root mass	O_2 transport	n
	g	mg O_2 /g/hr	
<u>Hydrocotyle umbellata</u>	0.02-0.05	3.95 ± 1.86	18
	0.06-0.12	2.49 ± 1.05	8
<u>Pistia stratiotes</u>	0.05-0.25	0.30 ± 0.13	10
<u>Eichhornia crassipes</u>	0.03-0.10	1.29 ± 1.18	10
	0.11-1.25	1.27 ± 0.61	10
	0.26-0.50	0.31 ± 0.11	8
	0.51-0.99	0.12 ± 0.14	4
<u>Sagittaria latifolia</u>	0.03-0.06	1.72 ± 0.87	15
	0.07-0.14	0.61 ± 0.22	3
<u>Typha</u> spp.	0.02-0.10	1.39 ± 1.49	4
	0.11-0.53	0.19 ± 0.15	14

2.4.2 Oil Clean-Up by Plants

The reported biological effects of repeated small oil spillages and chronic discharges range from localized and subtle to widespread and persistent (Dicks and Hartley, 1982). The vascular plants most commonly affected by oil pollution are saltmarsh and mangrove species (Baker, 1983). The study of oil spill clean-up techniques in Spartina alterniflora (Kiesling et al., 1988) shows that clipping followed by sorbent pad application on the

substrate removed 36 - 44% of added oil. Pilot plant trials with palm oil mill effluent have shown that water hyacinth can also be successfully grown in anaerobically digested liquor removing 97% oil and grease (John, 1984). Delaune et al. (1984) found a 31% reduction in sediment oil after clipping. In littoral areas, oiled substrate stripping is viewed as a radical clean-up approach.

2.5 The Application of Aquatic Macrophytes as Wastewater Treatment Systems

Because of their ability to remove pollutants from wastewater and the attendant low capital cost, many biotechnological wastewater treatment systems have been used to improve water quality such as those offered by high rate algal ponds (Shelef and Soeder, 1980), duckweed systems (Oron and Wildschut, 1984), water hyacinth systems (Weber and Tchobarioglous, 1985), land application (Abernathy et al., 1985), surface-flow reed beds (Alexander, 1985) and artificial wetland wastewater treatment systems (Seidel, 1976; Kickuth and Kaitzis, 1975). Among these alternatives, the root zone treatment systems and water hyacinth treatment systems are probably the most popular.

2.5.1 Use of Water Hyacinth in Wastewater Treatment

Of the known vascular aquatic plants, one of the most promising for industrial utilization is water hyacinth (Eichhornia crassipes) which, in view of its special characteristics, is an excellent material for a biological filtration system. The water hyacinth has been widely used for wastewater treatment in many countries because of its prolific growth rate and ability to remove pollutants from

wastewater. It was noted that two plants produce 300 units in 23 days and can cover an area of 4,000 km² in 8 months (Wolverton and McDonald, 1979). Biomass production is around 873 kg/ha/day matter (Wolverton and McDonald, 1975). Water hyacinth treatment systems can reduce COD, TSS, N, P, K, heavy metals, pesticides, petrol and oil (Gupta, 1980). John (1984) showed that water hyacinth can be successfully grown in anaerobically digested liquor removing 96% BOD, 87% COD, 96% suspended solids, 83% ammoniacal nitrogen and 97% oil and grease after 25 - 30 days (Table 2.8). It also removed more than 99% of indicative bacteria such as coliforms, Escherichia coli and Streptococci. Widyanto et al.(1983) suggested that water hyacinth can also absorb sodium, chlorine and sulphur from the wastewater. Xie et al.(1984) showed that the concentrations of Cu and Zn in sewage water decreased notably after absorption by water hyacinth for 15 days. Hardy and O'Keeffe (1985) showed that after exposure to Cd²⁺ for 4 hours, 300 mg dry weight rootmass can remove about 400 ug. Studies have shown that water hyacinth is suitable for treatment of industrial wastewater such as textile wastewater (Trivedy and Gudekar, 1987); natural rubber effluents and oil palm mill effluents (John, 1984) as well as pisciculture waters (Simeon and Silhol, 1987) and domestic sewage (Cloris and Araujo, 1987).

Santos et al. (1987) showed that, on a qualitative basis, the following assertions can be made to remove BOD:

a) Without water hyacinths in the reactor, the effluent would have a poor appearance due to the algae which would influence the BOD if not filtered. Even after filtering, the BOD could still increase due to the extracellular products usually excreted by algal metabolism.

b) The fixation of micro-organisms on the plant roots of water hyacinths increased the quantity of active micro-organisms inside the water.

c) It was observed by microscopic examination that the micro-organisms present on the plant roots were strictly aerobic. This aerobic zone could only be explained by the presence of the hyacinths. Once the aerobic zone is formed, there is an obvious increase in the decomposition rate due to aerobic metabolism, and a valuable contribution from the microfauna present on the roots (protozoa, rotifers, etc.) in the removal of carbonaceous matter.

Table 2.8: Effect of Growing Water Hyacinth In
Partially Digested Liquor (from John, 1984)

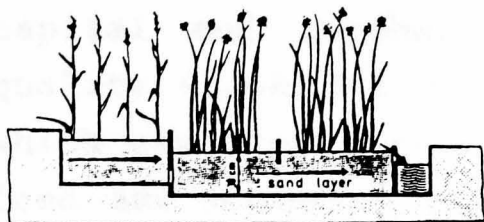
Parameter*	Raw	Hydraulic retention time (day)		
	effluent	10	20	25
pH	6.49	7.49	7.98	8.08(-)
BOD	4980	660	310	180(96.4)
COD	8850	2060	1500	120(87.3)
Suspended solids	3560	550	450	140(96.1)
Total solids	8210	5550	4690	4540(44.7)
Total nitrogen	285	140	80	65(77.2)
Ammoniacal nitrogen	120	55	25	20(83.3)
Oil and grease	660	25	30	20(97.0)

*All parameters except pH are expressed in mg/l.
The figures in brackets refer to percentage reduction.

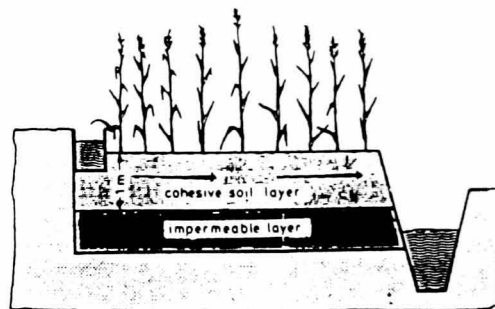
2.5.2 The Root Zone Biotechnology System of Wastewater Treatment

Emergent plants such as the common reed (Phragmites communis), common reedmace (Typha latifolia), narrow leaf reedmace (Typha angustifolia), bullrush (Schoenoplectus lacustris) etc. can grow rapidly in urban wetlands, are pollution tolerant, and produce a high biomass. In addition they can actively remove pollutants from wastewaters and supply oxygen to associated bacteria which proliferate in the aerobic conditions, thus creating stabilized organics in the effluent and nitrifying ammonia to nitrate (Lawson, 1985; Alexander and Wood, 1987).

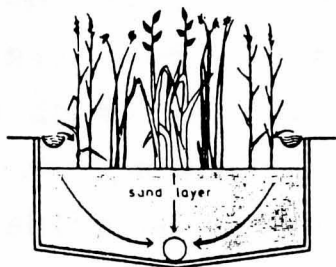
The macrophyte bed treatment systems approaches have been pioneered by both Seidel (1976) and Kickuth (1975). Seidel's process, known as the Krefeld system, utilizes Typha spp. planted in a gravel substratum (Fig. 2.3 a). Kickuth's process, known as the root zone system, utilizes Phragmites spp. planted in a soil substratum (Fig. 2.3 b). Both systems were developed for sewage treatment purposes with the removal of organics and ammonia from the wastewater as the prime consideration. Further variations on helophyte bed systems are shown in Fig. 2.3 c - e. The reed-bed and Root Zone Biotechnology (RZB) systems (Arthur, 1986) however have now become well established procedures in continental Europe and there are currently forty three Reed Bed Treatment Systems in operation at twenty seven sites within the UK.



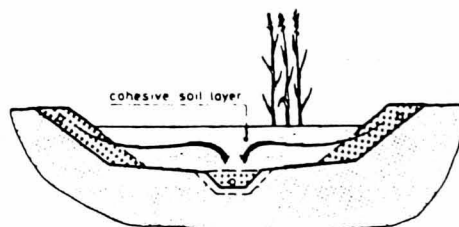
(a) Helophyte beds in series with gravel or sand as suggested by Seidel (1966, 1983)



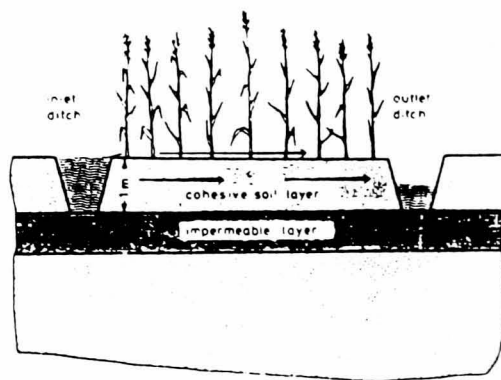
(b) Helophyte bed with cohesive soil as suggested by Kickuth (1980, 1984)



(c) Helophyte bed with gravel or sand, alternative solution to (a)



(d) Helophyte bed with cohesive soil, alternative solution to (b)



(e) Helophyte bed with cohesive soil, alternative solution to (b)

Fig.2.3 a - e Helophyte bed designs (from Bucksteeg, 1987)

It is an attractive technology, offering relatively low capital and revenue costs to attain water of a given quality (Loehr et al., 1979; Overcash and Pal, 1976) and which can cope with many types of effluent over and beyond food and domestic wastes (USEPA, 1977). RZB is ideal for small-scale/low-technology treatment situations, e.g. farm wastes, landfill leachate or treatment systems in developing countries. In the UK and Europe the use of reed beds as a controlled wetland for treating domestic effluent is currently attracting considerable interest. Studies have shown that aquatic macrophyte-based treatment systems offer a low-cost method of removing 80% to 90% of organic loading (BOD_5 , COD) and 85% TSS (Suspended Solids) from domestic wastewater (Boutin, 1987) and can remove 76% of COD and 82% of total-sulphur load from textile finishing effluent (Winter and Kickuth, 1989). Reed-beds can remove about 85% of nitrates and a large amount of phosphates as well as hydrocarbons, phenols, heavy metals and pathogenic bacteria. Reeds have the remarkable ability of absorbing oxygen through their above ground stomatal pores and of transporting it down to the root zone. This multiplies the oxidation value of the boundary surface some 40 times, hence the intensity of chemical and bacteriological processes in the root zone. The root zone treatment systems have been used for (1) raw sewage, (2) settled sewage and (3) biological treated sewage treatment (Bucksteeg, 1987). The pollutant removal capability of RZB systems has therefore been widely proven in wastewater effluents and sewage treatment particularly to meet tertiary treatment objectives at low cost in small communities (Cooper et al., 1989).

Despite these encouraging developments, the technology has its limitations and is certainly not a general panacea for

urban waste-water treatment. Consideration must be given to (a) the problems of public acceptance, (b) the potential contamination of ground and surface waters, (c) pathogens, and (d) contamination of the food chain (Bayes et al., 1989) as well as the potential volumes and loading rates required to be handled in urban areas.

A number of useful applications for water hyacinths have been investigated (Bates and Hentges, 1976; Widyanto et al., 1983; and Abdalla et al., 1987). These include schemes for harvesting to make compost and soil additives, to extract chlorophyll and carotene, to produce high-protein cattle food, to produce pulp, paper and fibre and more importantly to provide biogas such as methane as an energy source. The percent of methane in the biomass produced by anaerobic decomposition of water hyacinth contaminated with heavy metals is 91.1% while the non-contaminated plants produce biomass with 69.2% methane. One kilo dry matter produces 140 - 280 litre methane in 23 days (Wolverton and McDonald, 1975). The residual sludge may be utilized for recycling the metals (Table 2.9).

Table 2.9: Processes for the transformation of vascular aquatic plants into industrial products
(from Pinto et al., 1987)

Absorption and Removal of Substances	Plant Processes	Products
Removal of Heavy Metals and organic Substances from Industrial Effluents	Anaerobic Fermentation -----	Methane
	Residual ----- Metal ----- Sludge Extraction	Ag, Au, Cd, Hg, Pb, etc
Removal of Phosphates and Nitrates from Domestic Sewage	Aerobic Fermentation	Methane
	Residual -----	Fertilizers
	Dry Matter Processing --	Animal Food ----- Added to ration of cattle pigs, etc
		Human Food ----- Protein Supplementation (flour or bran)
	Composted -----	Fertilizer for Agriculture

Chapter 3: Locations and Methods

3.1 Field Study

3.1.1 Sampling Locations

Individual (Typha latifolia) plants and associated sediment and water samples for heavy metal analyses were collected at approximately bimonthly intervals between October 1988 and December 1989 from three sites in N London located within the London Borough of Barnet and having differing background levels of pollution (Fig. 3.1). These included a spring-fed pond situated in a country park estate (site 1), an ornamental pond receiving surface water discharges from the M1 motorway (site 2), and the Welsh Harp flood storage reservoir (site 3) on the lower reach of the Silk Stream feeder to the River Brent.

Site 1 (grid reference TQ 293973): This spring-fed pond, the Dew Pond, is situated within the Middlesex Polytechnic campus of Trent Park, on the 'green belt' fringe of North London (Fig. 3.2). The Dew Pond has an extensive bed of Typha latifolia whose annual leaf decomposition has contributed organic material to the benthal sediment. The absence of visitor pressure and management has led to the dense vegetation, including Salix sp, Crataegus sp and Rubus sp around the pond (Shutes, 1985). The pond receives some surface runoff from adjacent arable fields and meadows resulting in limited pollutant inputs. Samples were collected from the east margin of the pond (Plate 3.1).

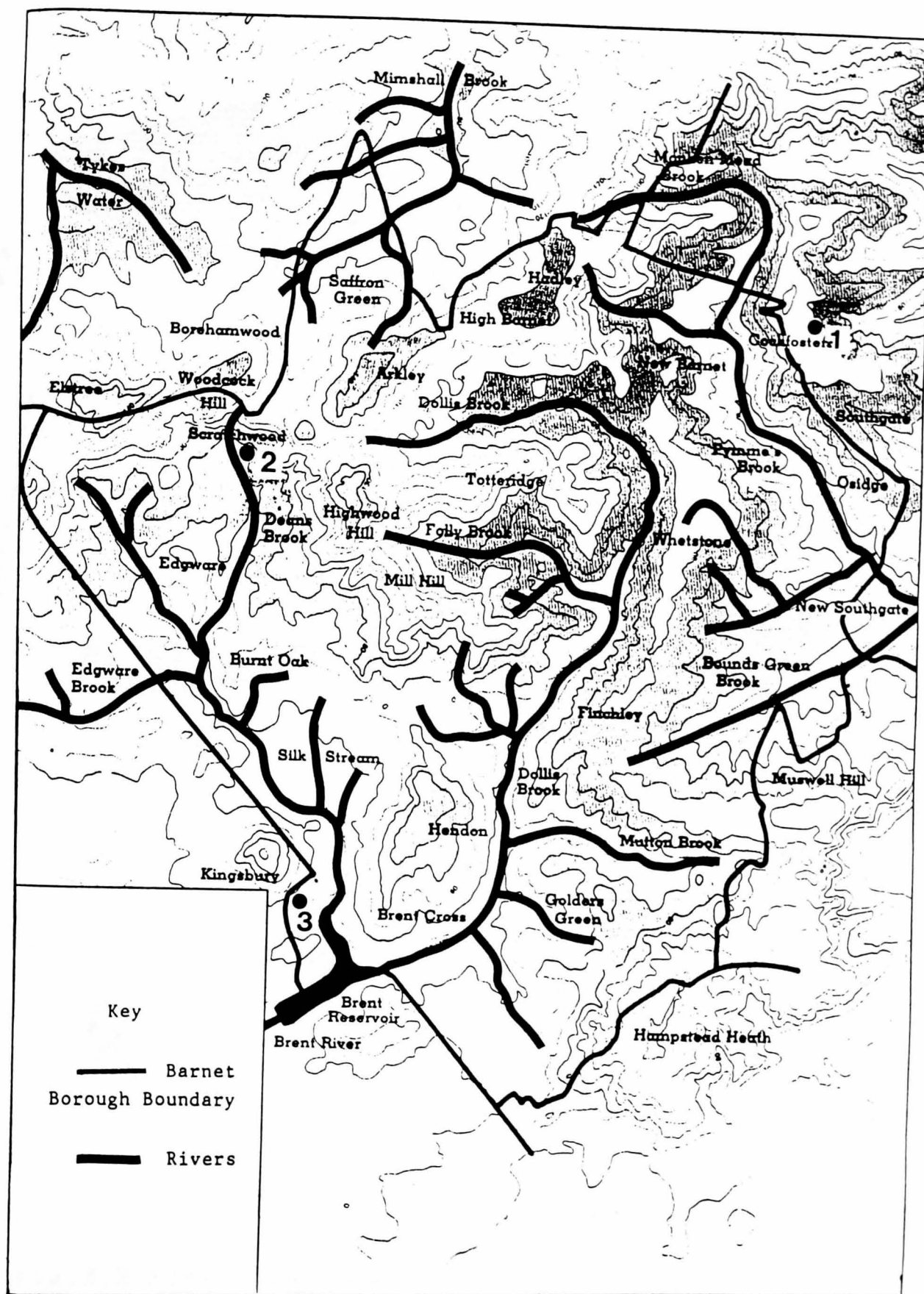


Fig.3.1 Sampling locations in North London (GLC, 1986)

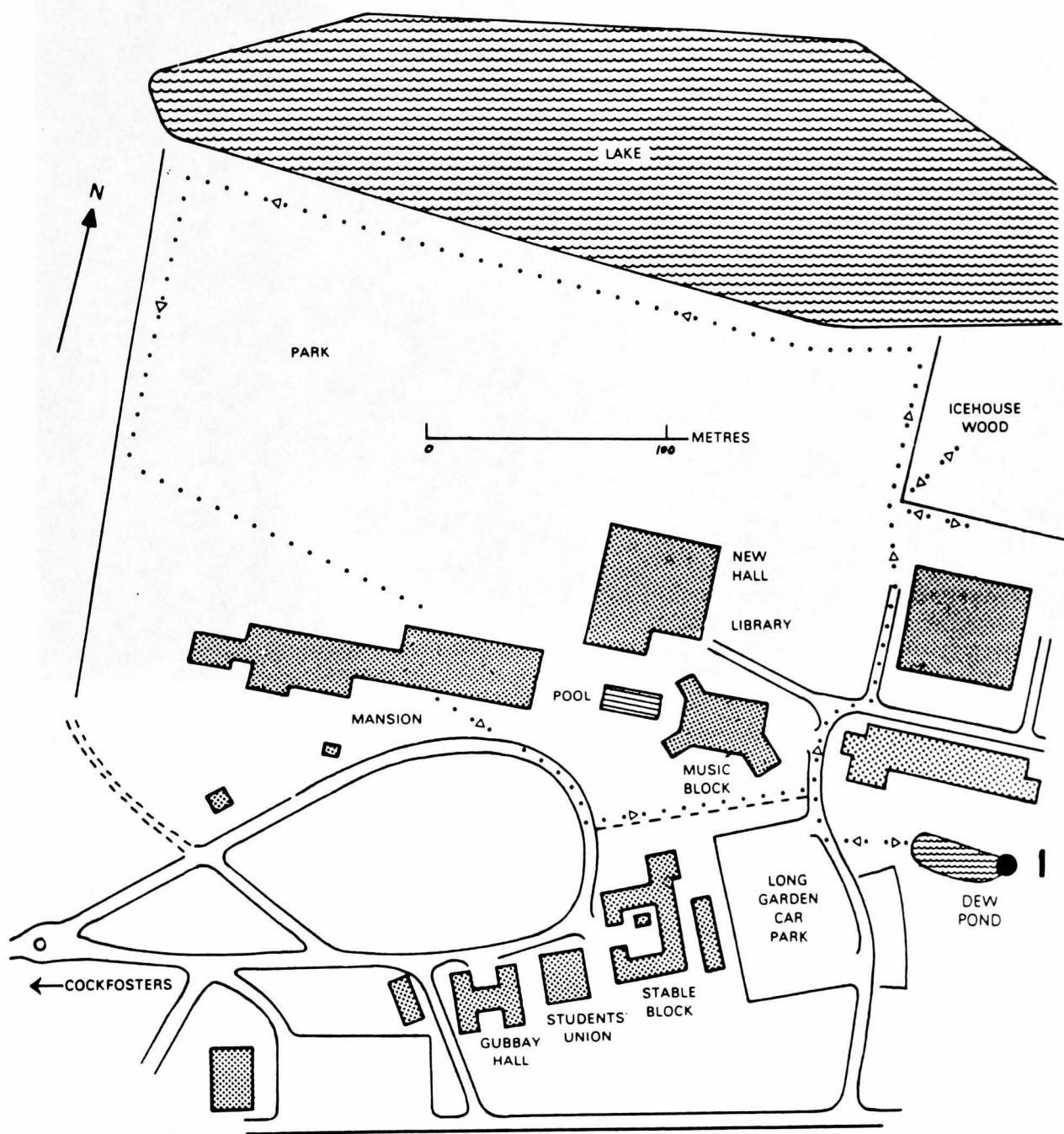


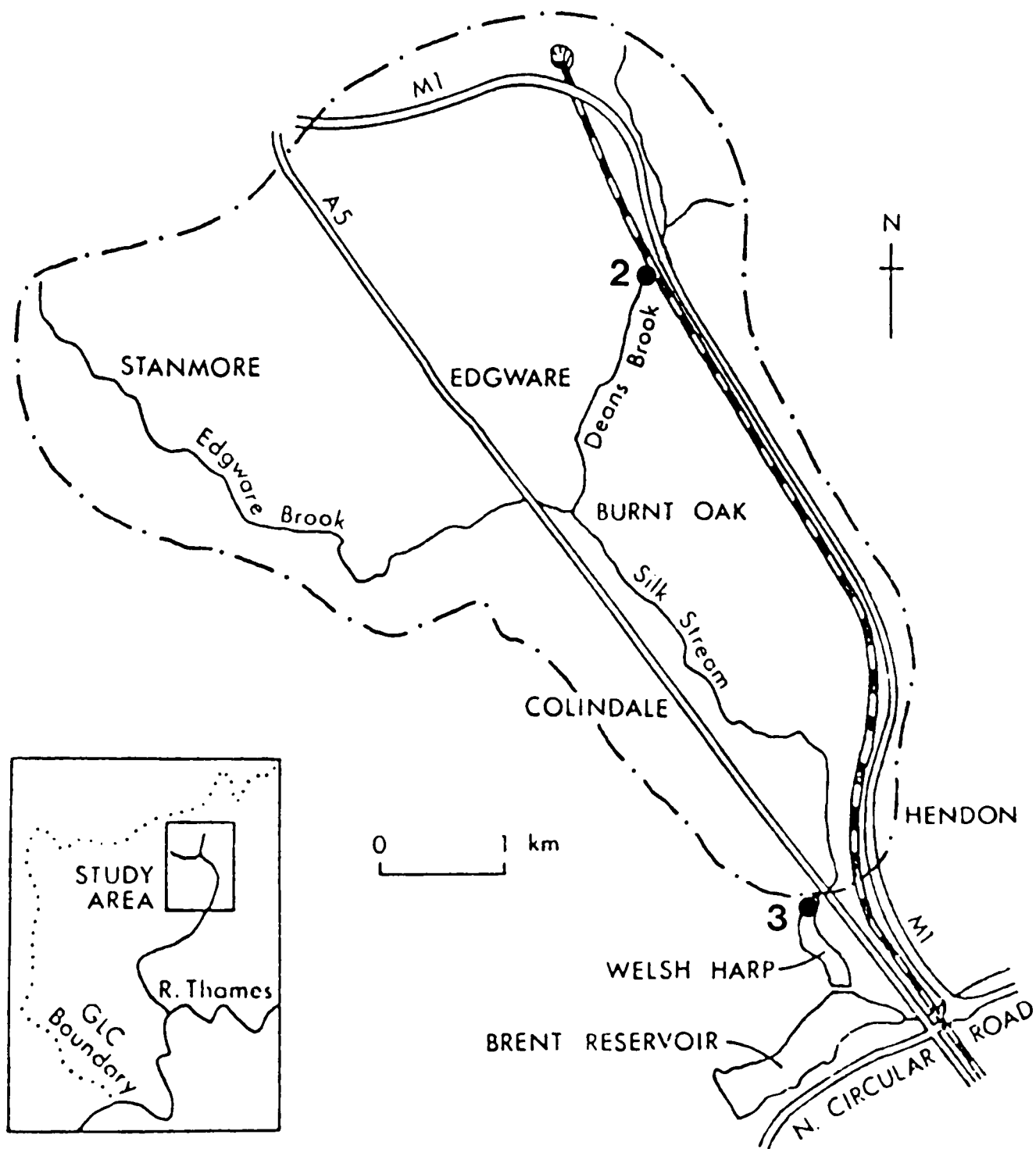
Fig.3.2 Site 1 The Dew Pond situated in Trent Park



Plate 3.1 Site 1 The Dew Pond situated in Trent Park

Site 2 (grid reference TQ 203931): This ornamental pond in Stonyfield Park is fed by the Dean's Brook a tributary of the Silk Stream, which receives surface runoff from the M1 motorway and other local road surfaces in NW London (Fig. 3.3). The Dean's Brook is one of the two tributaries of the Silk Stream and rises from a series of springs above a nearby golf course.

Samples were collected at the north west margin of the pond where the fringing vegetation is dominated by Typha latifolia (Plate 3.2).



----- Catchment Boundary

Fig.3.3 Sites 2 (Stonyfield Park) and 3 (Welsh Harp)

Site 3 (grid reference TQ 216882): The northern arm of the Welsh Harp reservoir in NW London comprised the third sampling location. This basin receives urban stormwater runoff via the Silk Stream which enters at its northern end (Fig 3.3). The catchment of the Welsh Harp is dominated by stormwater inputs from both combined and separate sewer systems, with 60% of the storm flow being derived from impermeable surface runoff (Hall, 1977). The Welsh Harp was built in the mid-nineteenth century by constructing a dam across the river Brent which rests on a bed of London Clay to create a total impoundment area of 190 acres. Until the early 1920s, the drainage basin was mainly rural but subsequent urbanisation has increased the percentage of impervious area in the catchment from 15% in 1929/30 to 25% in 1975/76 (Gavens et al., 1982). Urbanisation has increased at a rate of around one per cent of the land area each year and now comprises some 43% of the catchment. The reservoir is now a designated Site of Special Scientific Interest (SSSI), on account of its bird species and the marginal successional vegetation formed on the shallow areas of the Northern and Eastern Arms. There is an extensive area of wet woodland dominated by crack willow (Salix fragilis) surrounded by successional vegetation composed of greater and lesser reedmace (Typha latifolia and Typha angustifolia).

Samples were collected at the site which receives the Silk Stream at northern edge (Plate 3.3).



Plate 3.2 Site 2 The ornamental pond situated in Stonyfield Park



Plate 3.3 Site 3 The northern arm of the Welsh Harp reservoir

3.1.2 Sampling Methods

Large plastic containers, plastic bags and plastic sample bottles pre-washed with 2% nitric acid were prepared for plant, sediment and water samples respectively. Plants were dug out by spade and kept in the plastic containers. Surface sediments (top 2.5 cm) were obtained by grab samplers and stored in plastic bags. Water samples were collected in plastic bottles which were pre-washed with sample water and then filled to 2/3 capacity.

3.1.3 Laboratory Methods

Plants: On return to the laboratory, the plant samples were carefully washed to prevent tissue damage with tap water to remove any foreign material attached to the surface (leaves were washed with 2% diluted nitric acid) followed by deionised water to remove all traces of attached sediment. The plants were separated into three parts constituting leaf, rhizome and roots and each fraction was oven dried for 24h at 100°C. After grinding with a stainless steel mill, the oven dried plant material (2.5g) was digested for 24h with 10-15 ml concentrated nitric acid in a 100 ml pyrex beaker placed on a heated sand-bath and taken to dryness. The resulting residue was taken into solution with 2% hydrochloric acid and after filtering through a Whatman No.42 filter paper, the solution was made up to 100ml with deionised water. Duplicate samples were prepared for all plant fractions.

Sediments: Sediment samples were dried for 24h at 100°C in a hot-air oven and the dried sediment material (2.5g) was digested to dryness with 10-15ml concentrated nitric acid in a 100ml pyrex beaker placed on a heated sand-

bath. The resulting residue was taken into solution with 2% hydrochloric acid and after filtering through a Whatman No.42 filter paper, the solution was made up to 100ml with deionised water. Duplicate samples were prepared for all sediment fractions.

Analytical Techniques for Plants and Soil: The concentrations of Cu, Pb, Zn and Cd in the extracted samples were measured by means of a Philips SP9 Atomic Absorption Spectrophotometer (AAS) in the flame atomisation mode (Plate 3.4).

Standards were prepared from Spectrosol standard solutions (1000 ppm). A 100 ppm stock was used to prepare standards of 1, 2, 5, 10 ppm of Cu, Pb, Zn, Cd for calibration. The SP9 Computer was set to a fixed ratio programme of 4 standards giving automatic calibration with curvature correction.

The AAS was used with the following operating conditions:-

- (i) deuterium B/C correction
- (ii) 0.2 nm bandpass setting
- (iii) air/acetylene flame
- (iv) 10 sec. integration time

Each lamp was allowed an initial heating up time of 10 minutes and a little less than maximum current was used in order to give greater sensitivity. Cu, Pb, Zn, and Cd lamps were set at wavelengths of 342.8 nm, 217.0 nm, 213.9 nm and 228.8 nm respectively.

A blank of deionised water was used before the standards were measured in ascending order of concentration.

Once the system was calibrated, sample concentrations were measured and converted to parts per million (ppm) expressed on a dry weight basis as plant or sediment.

Water: Water samples (50ml) were heated to dryness on a hot sand-bath after the addition of 2ml concentrated nitric acid. The residue was taken into solution with 2% dilute nitric acid and after filtration, deionised water was added to give a final volume of 50ml. Duplicate samples were prepared for each water sample. The concentrations of heavy metals (Cu, Pb, Zn and Cd) in the water samples were measured by means of a Princeton Applied Research Model 384 Polarographic Analyser (Plate 3.5) using the standard addition method.

Analytical Techniques for Water: The operating procedure using the instrument parameters shown in Table 3.1 were follows:

(1) 7ml of the acid digested water sample and 3ml of sodium acetate buffer (2M) were placed in the sample cell and analysed by the Anodic Stripping Voltammetry (ASV) technique.

Table 3.1: Parameters used for Model 384
Polarographic Analyser

Parameters			
Initial E	-1.200 V	Equilibration time	30 seconds
Final E	0.000 V	Pulse height	0.050 V
Purge time	240 seconds	Replications	1
Drop time	1.0 seconds	Deposition time	60 second
Scan increment	2 MV	Conditioning time	0 seconds



Plate 3.4 Philips SP9 Atomic Absorption Spectrophotometer used for sediment and plant analysis



Plate 3.5 Model 384 Polarographic Analyser used for water analysis

(2) The above solution was spiked with 0.02 ml of a mixed 10 ppm Cu, Pb, Zn and Cd solution and the ASV analysis repeated.

(3) Procedure (2) was repeated.

The results were printed as graphs and after the calculation the concentration was expressed as parts per billion (ppb).

3.1.4 Biomass Measurement

The Typha latifolia plants in a 0.5 m square quadrat were counted and completely removed from site 1 on 3 October, 1989. The numbers of shoots within each quadrat were counted. Plants were taken back to the laboratory and washed with tap water. The roots, rhizome and leaves of the plants were separated and their wet weights determined. The plant tissues were oven dried for 24 hours and their dry weights measured.

3.2 Greenhouse Experiments

Greenhouse experiments with Typha latifolia and Juncus effusus were carried out at the Middlesex Polytechnic Centre for Urban Pollution Research at Enfield and treated as follows.

3.2.1 Short Term Heavy Metal Dosing Experiments

Typha plants were collected on five occasions from the relatively unpolluted Dew Pond, at Trent Park (site 1) between 19 January, 1989 and 25 April, 1989. The plants were placed in 3 plastic tanks (35 cm high and 30 cm diameter) containing Irish Moss Peat. The background

concentrations of heavy metals (Cu, Pb, Zn and Cd) in the peat were determined so as to assess any bias from contaminated peat. Irish Moss Peat was chosen because of its presumed low background metal levels (Cu, 2.0; Pb, 11.2; Zn, 19.2 and Cd, <0.01 mg/kg). 2 dosage tanks and 1 control tank were used, each containing three to five plants.

The plants were dosed with 2 mg/l and 5 mg/l concentrations of Cu, Pb, Zn and Cd as their nitrate salts using 7.5 litres of the appropriate standard solution. The same amount of tap water was added to the control tank. After 10 days, plant, peat and water samples were collected from the 3 tanks and were analysed for metals by the methods outlined above. The leaf, rhizome and root tissues of Typha latifolia were analysed separately.

3.2.2 Long Term Heavy Metal Dosing Experiments

Long term heavy metal dosing experiments were also carried out over a period of eight weeks with both Typha and Juncus plants.

Dosing experiment with Typha latifolia: This experiment was carried out between 26 June, 1989 and 21 August 1989 using plants collected from site 1. Four dosage tanks and 1 control tank were used, each containing three to five plants.

Plants were dosed with 10 mg/l concentrations of Cu, Pb, Zn and Cd as their nitrate salts. 7.5 litres of 10 mg/l concentrations of mixed Cu, Pb, Zn and Cd solution was added to each of the dosage tanks while the same volume

of tap water was added to the control tank. One plant and associated peat samples were removed each week from the dosage tanks and analysed by the methods described previously (plant and associated peat samples from the control tank were analysed after 8 weeks). The peat samples were obtained from both the surface and bottom layers within the containers. The leaf, rhizome and root of Typha latifolia were analysed separately. The experiment was continued over 8 weeks and the plants in both dosage and control tanks were watered with beaker (water was added to sediment) approximately twice weekly (Table 3.2) with 10 mg/l concentrations of mixed Cu, Pb, Zn and Cd solution.

Table 3.2: Dosing Frequency and Water Volume
Used for Dosing Experiment with Typha

Date	Water* added (l)	Date	Water* added (l)
26/6	7.5	25/7	2.5
30/6	2.5	27/7	1.5
3/7	2.5	30/7	1.7
7/7	2.5	4/8	1.7
11/7	2.5	9/8	2.5
14/7	1.7	11/8	2.5
18/7	1.7	13/8	2.5
21/7	2.5	18/8	2.5

* mixed 10 ppm Cu, Pb, Zn and Cd solution

Dosing experiment with Juncus effusus: This experiment was carried out between 3 October 1989 and 29 November 1989 using plants collected from site 1. Four dosage

tanks and 1 control tank were used, each containing ten to fifteen plants.

Plants were dosed with 10 mg/l concentrations of Cu, Pb, Zn and Cd as their nitrate salts. 7.5 litres of 10 mg/l concentrations of mixed Cu, Pb, Zn and Cd solution was added to each of the dosage tanks while the same volume of tap water was added to the control tank. Five plants and associated peat samples were removed each week from the dosage tanks and analysed by the methods described previously (plant and associated peat samples from control tank were analysed after 8 weeks). The peat samples were obtained from both the surface and bottom layers within the containers. The leaf and root of Juncus were analysed separately. The experiment was continued over 8 weeks and the plants in both dosage and control tanks were watered approximately once weekly (Table 3.3) with 10 mg/l concentrations of mixed Cu, Pb, Zn and Cd solution.

Table 3.3: Dosing Frequency and Water Volume
Used for Dosing Experiment with Juncus

Date	Water* added (l)	Date	Water* added (l)
3/10	7.5	31/10	1.7
10/10	2.5	7/11	1.7
15/10	2.5	14/11	2.5
20/10	1.7	22/11	2.5
26/10	2.5	29/11	2.5

* mixed 10 ppm Cu, Pb, Zn and Cd solution

Dosing experiment with peat: An additional experiment was carried out using a tank containing only Irish Moss Peat to assess the uptake metal capacity of the peat in the absence of plants. The tank was dosed approximately twice weekly (Table 3.4) with 10 mg/l concentrations of mixed Pb, Zn, Cu and Cd solution as their nitrate salts over 8 weeks period. The peat samples were obtained from both the surface and bottom layers within the container and analysed by the methods described previously.

Table 3.4: Dosing Frequency and Water Volume
Used for Dosing Experiment with Peat

Date	Water* added (l)	Date	Water* added (l)
18/1	7.5	17/2	2.5
23/1	2.5	20/2	2.5
26/1	5.0	24/2	2.5
2/2	2.5	30/2	5.0
8/2	2.5	5/3	2.5
13/2	5.0	10/3	2.5

* mixed 10 ppm Cu, Pb, Zn and Cd solution

Chapter 4: Dosing Experiments

4.1 Introduction

Aquatic macrophytes are able to accumulate heavy metals to high levels (Chigbo et al., 1982; Muramoto and Oki, 1983; Sridhar, 1986; Blake et al., 1987), therefore, over the last 15 years these macrophytes have been studied extensively with regard to their use as indicators of pollution (Beeftink et al., 1982; Taylor and Crowder, 1983) and as water purifiers (Seidel et al., 1978; Black and DuBois, 1982). The emergent macrophyte Typha latifolia in particular grows rapidly and possesses a large capacity for assimilating mineral compounds (Black et al., 1984). A genetically based tolerance has been proposed for this species by Antonovics (1975). However, the ability of Typha populations originating from both contaminated and uncontaminated locations to tolerate high metal loads implies a constitutional tolerance (McNaughton et al., 1974; Taylor and Crowder, 1983). Typha has therefore been selected for studying metal uptake in this investigation. The aim of this study was thus to determine metal uptake by sediments and plants under greenhouse conditions by Typha latifolia. The metal uptake ability of Typha has been compared with Juncus effusus.

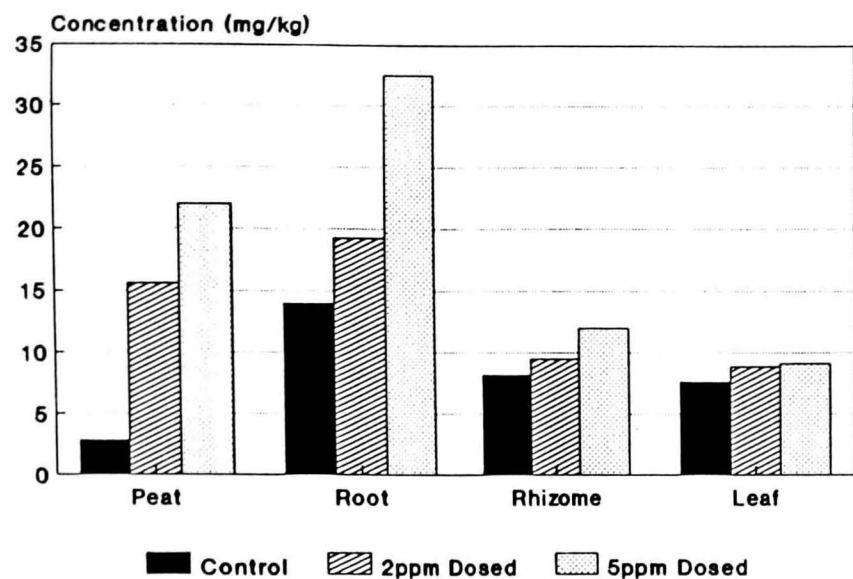
In controlled greenhouse-based phytoassay experiments Typha and Juncus were transplanted to Irish moss peat from site 1 and dosed with set metal concentrations to determine both uptake rates and maximum storage capabilities.

4.2 Discussion of the results

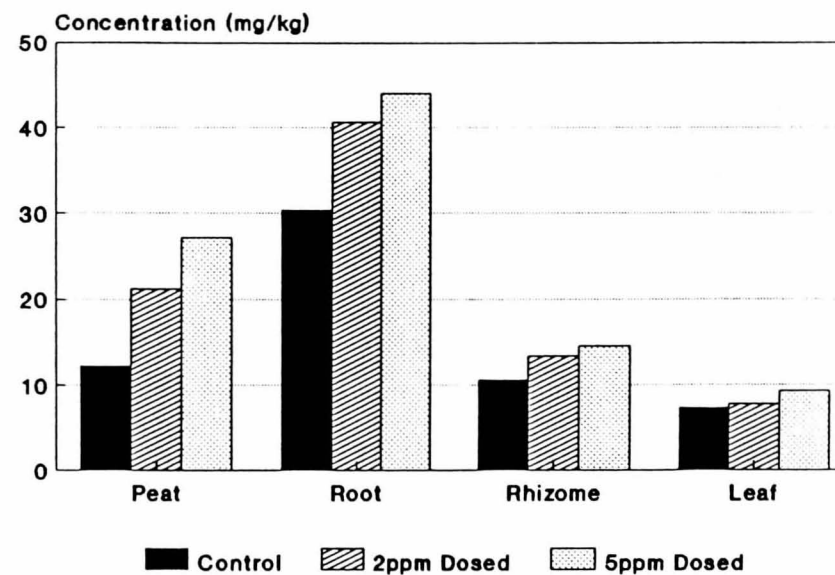
4.2.1 Short term dosing experiments

The short term greenhouse based metal uptake experiments were carried out over a ten day period (Chapter 3). Typha latifolia plants collected from site 1 were used for these tests and did not suffer any apparent toxic effects over the duration of the test. The results of metal uptake by peat and Typha plants over this time period are shown in Fig. 4.1. After 10 days, the metal levels in peat, Typha root, rhizome and leaf increased 72.1 %, 45.3 %, 36.7 % and 26.3 % respectively in 5 ppm dosed tests and when the dosing water concentrations increase, the concentration difference between peat, root, rhizome and leaf also increased.

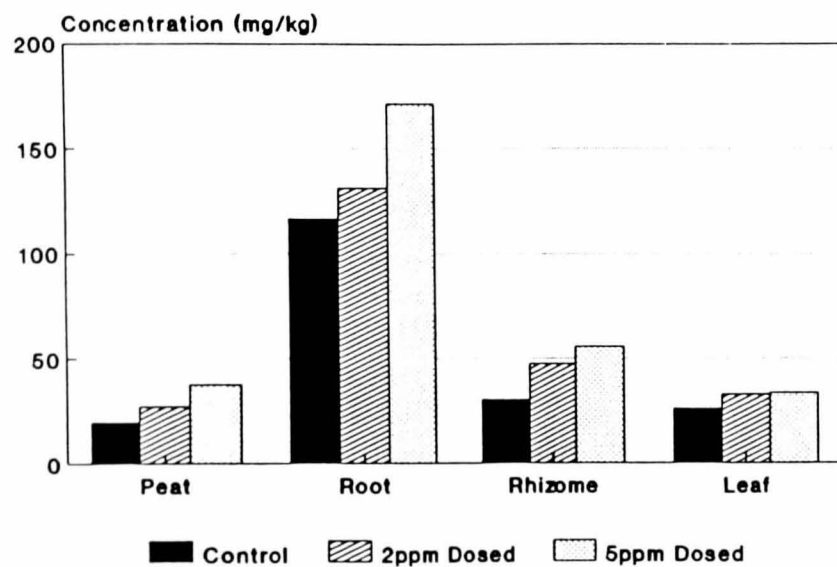
Peat metal levels were considerably enhanced over the 10 day experimental period with this being most noticeable for 5 ppm dosed peat. Metal levels increased from a base of 0.5 to 34.8, 2.8 to 22.0, 12.2 to 27.2 and 17.8 to 37.6 mg/kg for Cd, Cu, Pb and Zn respectively. Cu and Cd levels in dosed peat exhibit much higher concentration ratio increases compared to Pb and Zn over the 10 day dosing period. The ratios of the final concentrations achieved by the 5 ppm dose relative to the control peat are 68.0:1, 7.9:1, 2.2:1 and 1.9:1 for Cd, Cu, Pb and Zn, respectively (Table 4.1). Cd levels in 2 ppm and 5 ppm dosed peat after 10 days experiment exceeded the Cd levels observed in plant root tissues, whereas Cu, Pb and Zn levels in the dosed peat remained lower than the metal levels achieved in the plant root tissue.



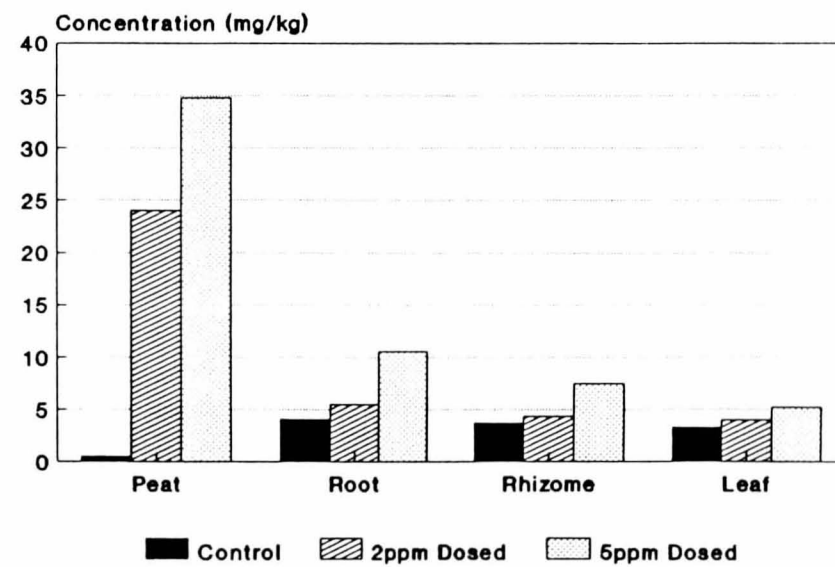
(a) Copper



(b) Lead



(c) Zinc



(d) Cadmium

Dosing time 10 days

Fig.4.1 Heavy metal levels in peat and Typha tissues

Metal levels in the plant root, rhizome and leaf tissues increased consistently over the 10 days dosing experiment with the highest levels being demonstrated by the 5 ppm dosed plant root tissues. The highest concentrations of all the metals were found in plant root tissues with a progressive decrease to rhizome and leaf tissues.

Table 4.1: Ratios of peat, root, rhizome and leaf metal levels relative to control peat

		5 ppm dosed*	2 ppm dosed*	Control
		(mg/kg)		
Peat	Cu	7.9	5.6	1 (2.8)
	Pb	2.2	1.7	1 (12.2)
	Zn	1.9	1.4	1 (19.8)
	Cd	68.0	48.0	1 (0.5)
Root	Cu	2.3	1.4	1 (14.0)
	Pb	1.4	1.3	1 (30.4)
	Zn	1.5	1.2	1 (117.0)
	Cd	2.6	1.4	1 (4.1)
Rhizome	Cu	1.3	1.2	1 (8.1)
	Pb	1.4	1.3	1 (10.6)
	Zn	1.8	1.6	1 (30.4)
	Cd	2.0	1.2	1 (3.7)
Leaf	Cu	1.2	1.2	1 (7.6)
	Pb	1.5	1.1	1 (6.3)
	Zn	1.3	1.3	1 (26.0)
	Cd	1.6	1.6	1 (3.3)

* dosing time 10 days

In parallel to the metal elevation in peat, Cu and Cd levels in 5 ppm dosed plant roots are more enhanced than Pb and Zn, whereas the ratios of dosed to control rhizome and leaf for all metals are relatively stable. A similar rapid uptake of Cd and Cu has been shown by experiments with Callitriche platycarpa and Elodea nuttallii over 14 days (Van der Werff and Pruyt, 1982).

4.2.2 Long term dosing experiments

In order to examine metal uptake by sediment and plant over the longer term, the dosing experiments were carried out over an extended eight week period of time with both Typha latifolia and Juncus effusus.

4.2.2.1 Long term dosing experiments with Typha latifolia

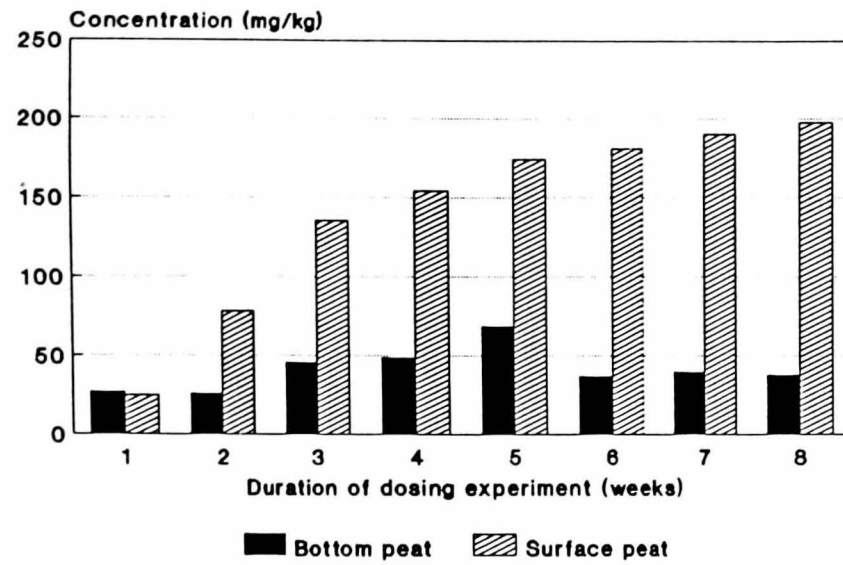
In the greenhouse-based metal uptake experiments carried out over an eight week period with Typha latifolia, the dosing metal levels of 10 mg/l were considerably enhanced in comparison to the average soluble metal concentrations found at the most polluted field site (site 3; Cd 8.9 ug/l; Cu, 53.4 ug/l; Pb, 36.2 ug/l and Zn, 136.6 ug/l). These long term, high dosing level tests are not entirely unrealistic in comparison with environmental field situations where the observed sediment metal concentrations (site 3; Cd, 12.4; Cu, 220.1; Pb, 841.2 and Zn, 778.9 mg/kg) are considerably higher for Pb and Zn compared to those monitored in the Irish Moss peat during the duration of the experiment.

The surface peat metal levels demonstrated large enhancements over the eight week experimental period with linear increases from < 0.01 to 286, 2 to 187, 11.2

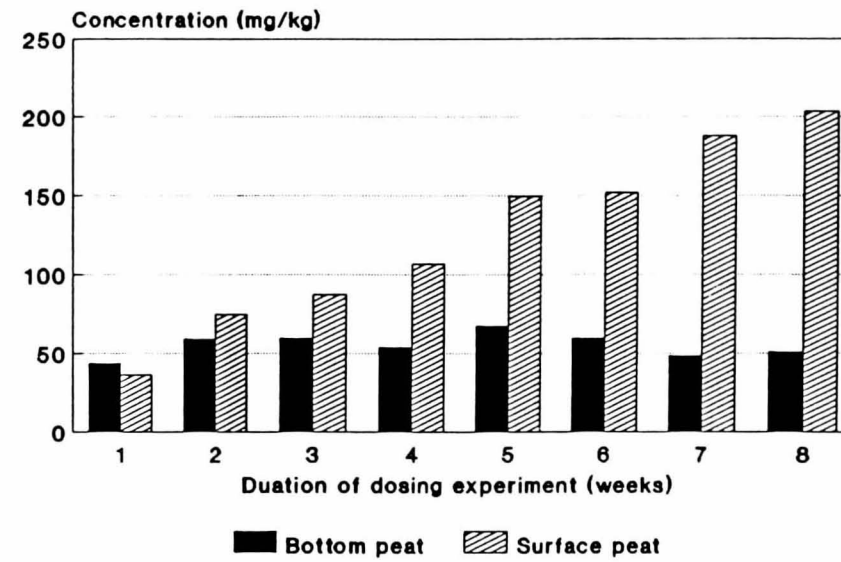
to 168.0 and 19.2 to 294.8 mg/kg for Cd, Cu, Pb and Zn respectively. The bottom peat metal levels remained relatively stable although with some random fluctuations (Fig.4.2). As the dosing time increases, the metal level difference between surface and bottom peat increases. The surface metal levels are up to 4.8 times higher than those of the basal peat. (Table 4.2). These results agree with a previous study by Blake et al. (1987) which showed a major proportion of added Zn was found in the surface sediment and the top 0.2 cm sediment layer had a concentration 2.5 times higher than the sediment Zn concentration found at a depth of 1.0 cm. In the present study, surface Zn concentration is 4.3 times higher than the bottom peat at a depth of 15 cm at the end of the 8 week experimental period (Table 4.2).

Table 4.2: Metal concentration ratios of surface to bottom peat during experiment with Typha

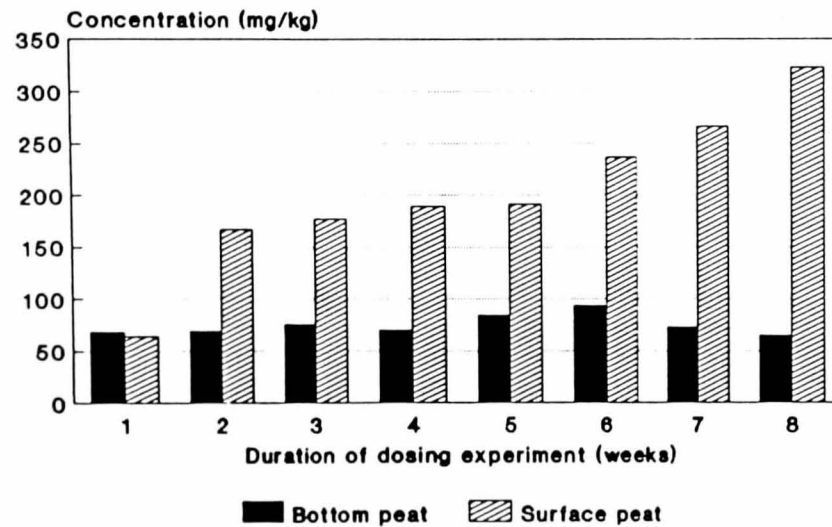
	1-2	3-4	5-6	7-8 (dosing weeks)
Cu	2.0	3.1	3.5	4.8
Pb	1.1	1.5	2.3	3.4
Zn	1.7	3.0	2.4	4.3
Cd	1.2	1.7	1.8	4.1



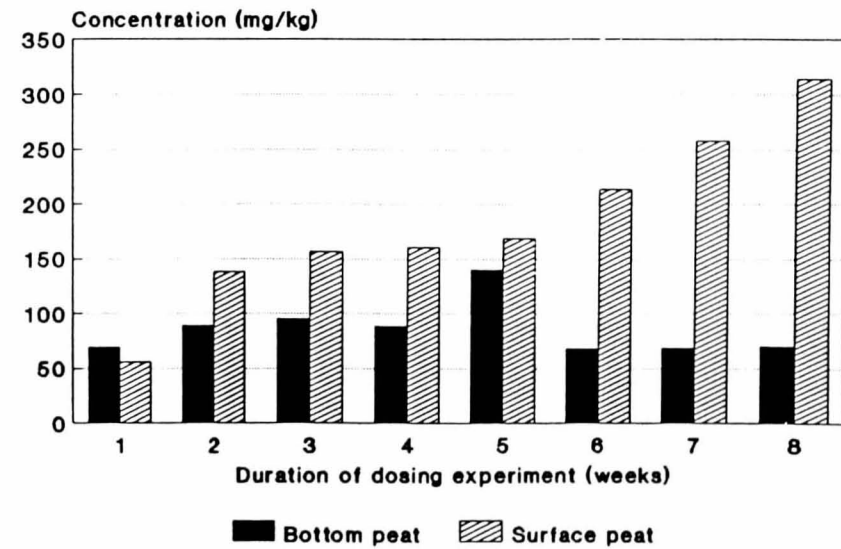
(a) Copper



(b) Lead



(c) Zinc



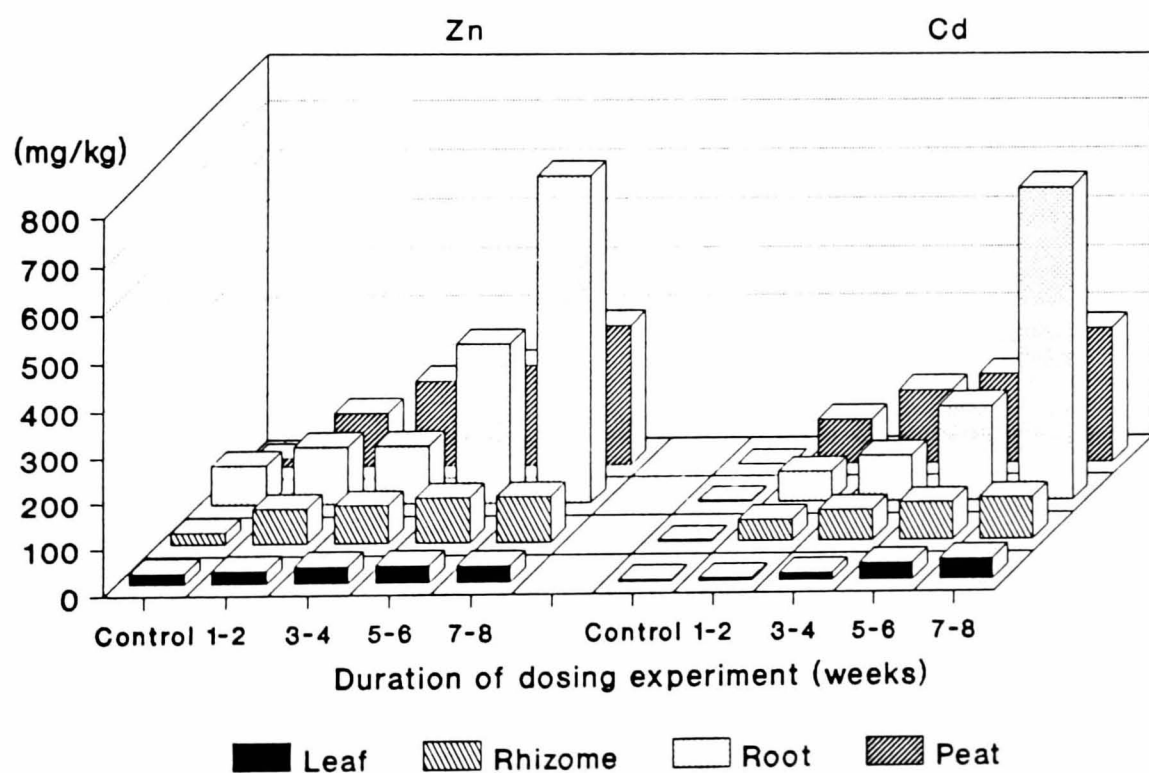
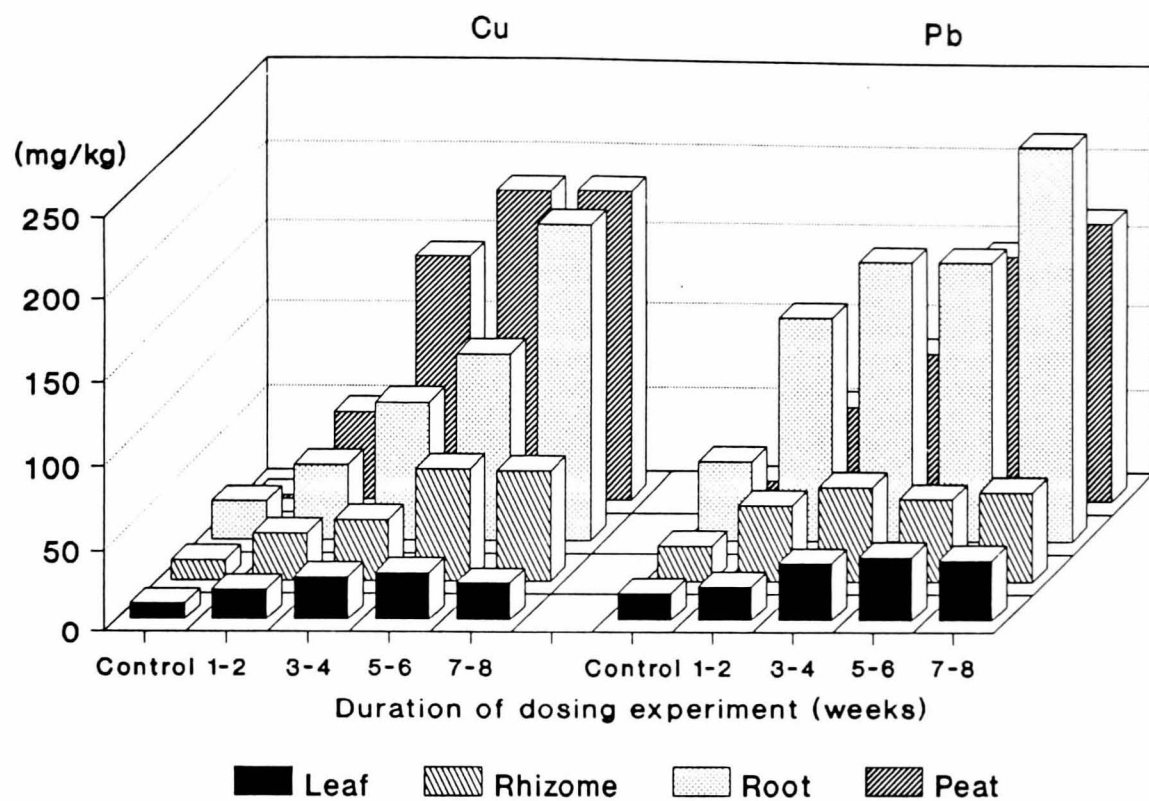
(d) Cadmium

Dosing concentration 10 ppm
Dosing time 3/10/1989 - 29/11/1989

Fig. 4.2 Metal level variations in surface and bottom peat samples during Typha latifolia 8 week dosing experiments

The surface sediment increases are most closely paralleled by the metal uptake patterns of Typha root tissue with the rhizome showing a reduced metal affinity and the leaf material being clearly the least important site with regard to metal concentrations (Fig. 4.3). Only in the case of Pb, does the leaf tissue eventually achieve comparable metal levels to the rhizome. This observation essentially agrees with that reported by Blake et al. (1987) in which the highest concentrations of Zn were found in the roots of Typha latifolia and were in the order of roots>rhizome>leaf tissue.

The observed peat, Typha root, rhizome and leaf metal uptake data over the eight week time period of the dosing experiment were analysed by statistical regression methods. The resulting equations are shown in Fig.4.4. The regression analysis shows that Typha leaf and rhizome metal uptake patterns are all linear (except for leaf Cd uptake which is exponential). The b values in the equations of leaf metal uptake are lower, so the patterns are more consistent. The root metal uptake patterns are less consistent, with Zn and Cd both exhibiting exponential increases with time and final values approach 700 mg/kg. These metals show the greatest metal enhancement in the root compared to the rhizome and leaf system but only after prolonged exposure.



Dosing concentration 10 ppm
Dosing time 26/6/1989 - 21/8/1989

Fig.4.3 Temporal increases in peat and Typha tissue metal concentrations during 8 week dosing experiment

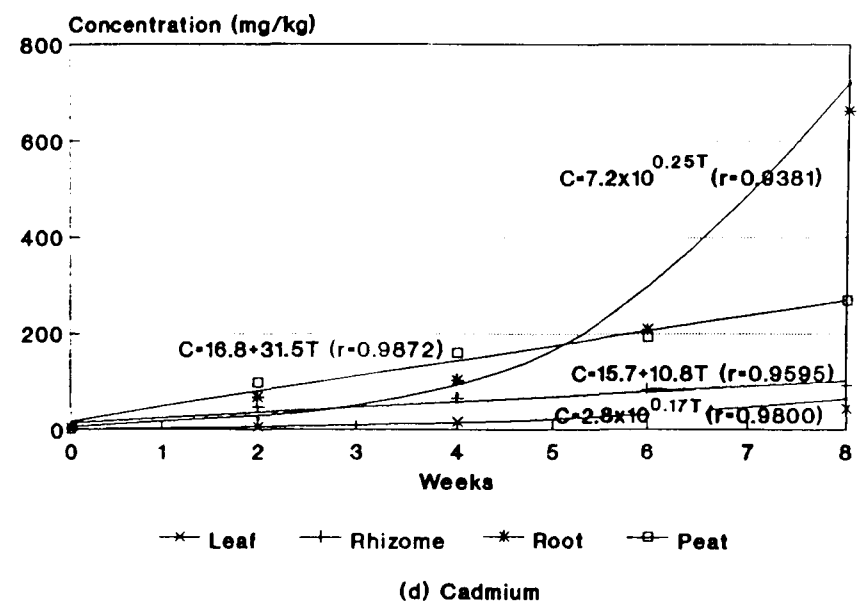
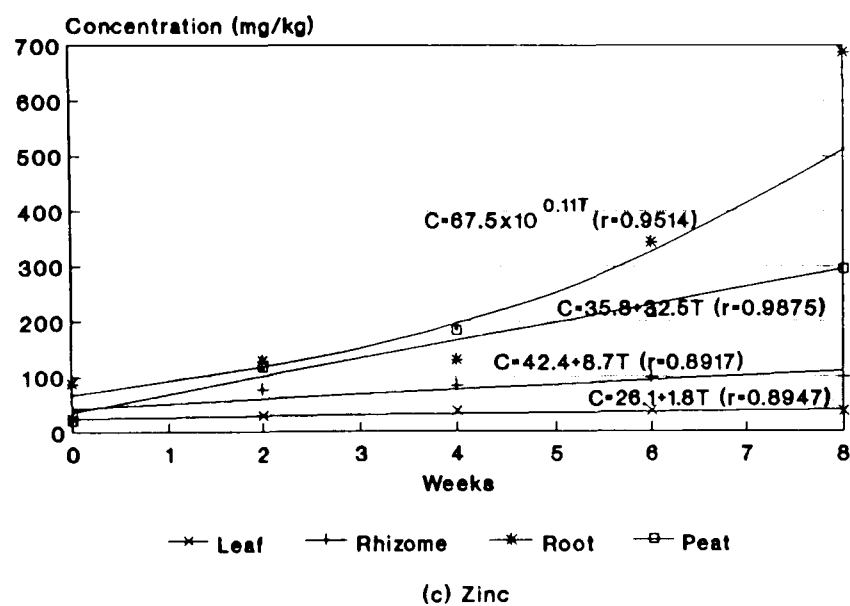
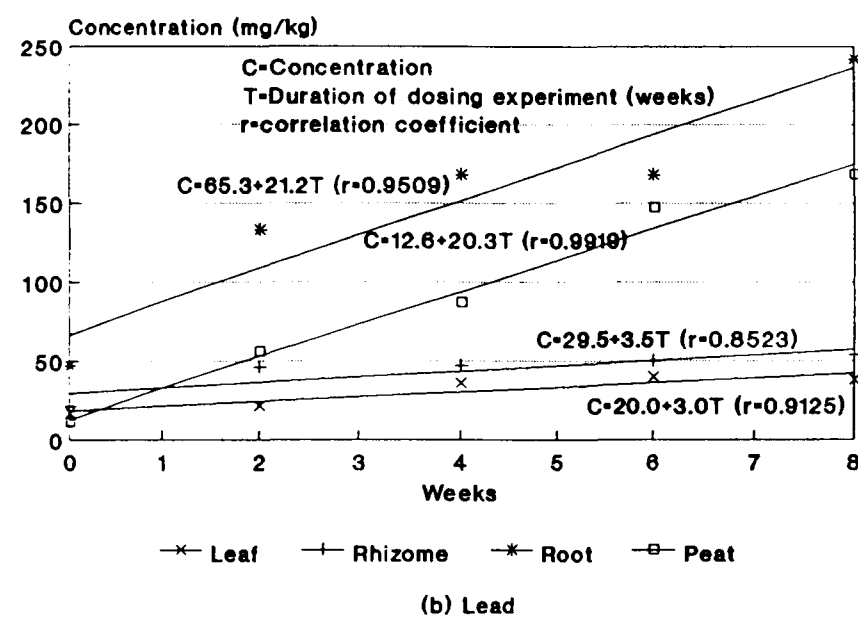
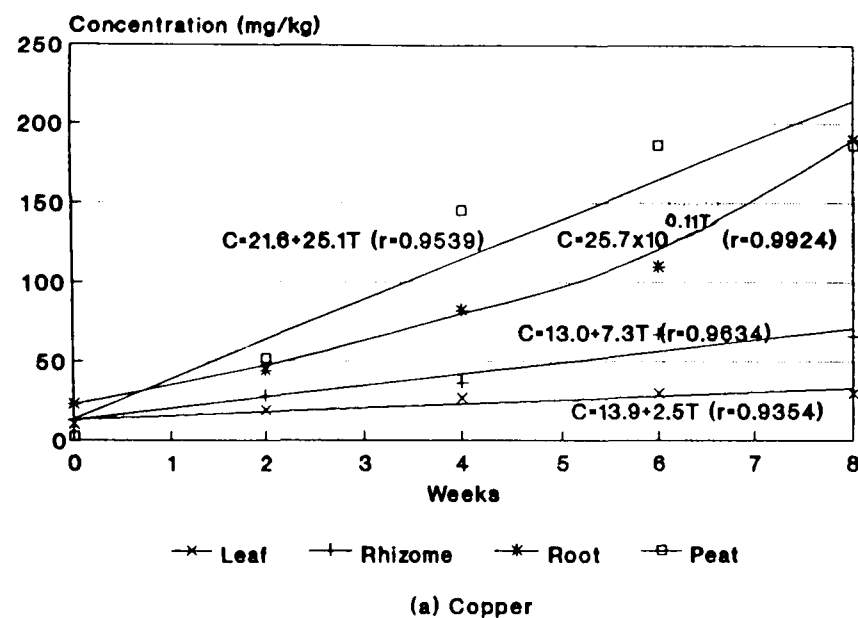


Fig.4.4 Regression equations and graphs for metal uptake by peat and Typha tissues during 8 week dosing experiments

The final root Cd and Zn levels are considerably elevated over those in the surface peat which corresponds to their established existence in urban waters as predominantly soluble bioavailable metal ions or weak complexes (Revitt and Morrison, 1987). However, the delay in uptake which can be seen from Fig.4.4 would appear to be due to the superimposed influence of sediment-water interactions which only release the bioavailable metals for plant uptake once a particular sediment saturation level has been reached. This saturation appears to be achieved after 4-5 weeks resulting in enhanced root uptake at this stage. Previous experiments with Typha latifolia (Blake et al, 1987) have shown Zn levels in the plant root tissue reaching 1400 mg/kg after dosing with 10 ppm solution for one month.

In comparison to Cd and Zn, Cu and Pb root tissue concentrations demonstrate lower accumulative metal levels after eight weeks reaching values of 190 and 242 mg/kg respectively. Cu root tissue concentrations exhibit a less exaggerated exponential increase with time whereas peat Cu concentrations achieve a value of 187 mg/kg. This is consistent with the established affinity of Cu with the humic and fulvic acids associated with soils (Luoma and Davis, 1983). In contrast to the other metals, Pb root tissue concentrations exhibit a linear increase showed less affinity of Pb compared to Zn and Cd. This is consistent with the previous studies of Miller et al. (1979) and Chigbo et al. (1982).

However, the metal root level ratios of dosed to control plant after 8 weeks are 200.8, 8.3, 7.8 and 5.1 for Cd, Cu, Zn and Pb respectively. This suggests that the uptake ability is in the order of Cd>Cu>Zn>Pb.

Interpretation of the variations in metal concentration suggests that the root to rhizome transfer mechanism, assuming negligible direct absorption, is most efficient for Cu followed by Pb with Cd and Zn and shows preferential metal accumulation within the root zone. Previous studies have demonstrated that up to 75% of added Zn tends to be deposited in Typha roots (Blake et al., 1987) and about 80% of the total Cd uptake was also found in Eichhornia crassipes (water hyacinth) roots by Blake et al. (1987).

Metal concentration values of Typha can be converted to tissue loadings using the dry weight biomass tissue ratios for Typha collected from Site 1 (root: rhizome: leaf = 1:18:14, see Chapter, 7); the metal distributions are as shown in Table 4.3. The metal loading data clearly indicate that the major metal bioaccumulation target area in Typha is the rhizome. It is interesting that for all metals, the root and leaf tissues show similar overall metal bioaccumulation loads. The leaf metal yields do have important implications for pollution control in terms of harvesting of the emergent foliage.

Table 4.3: Metal Loads in Typha Tissue for Dosing Experiment*

	Root	Rhizome	Leaf
Cd	662	1669	613
Cu	190	1188	329
Pb	242	976	532
Zn	689	1800	512

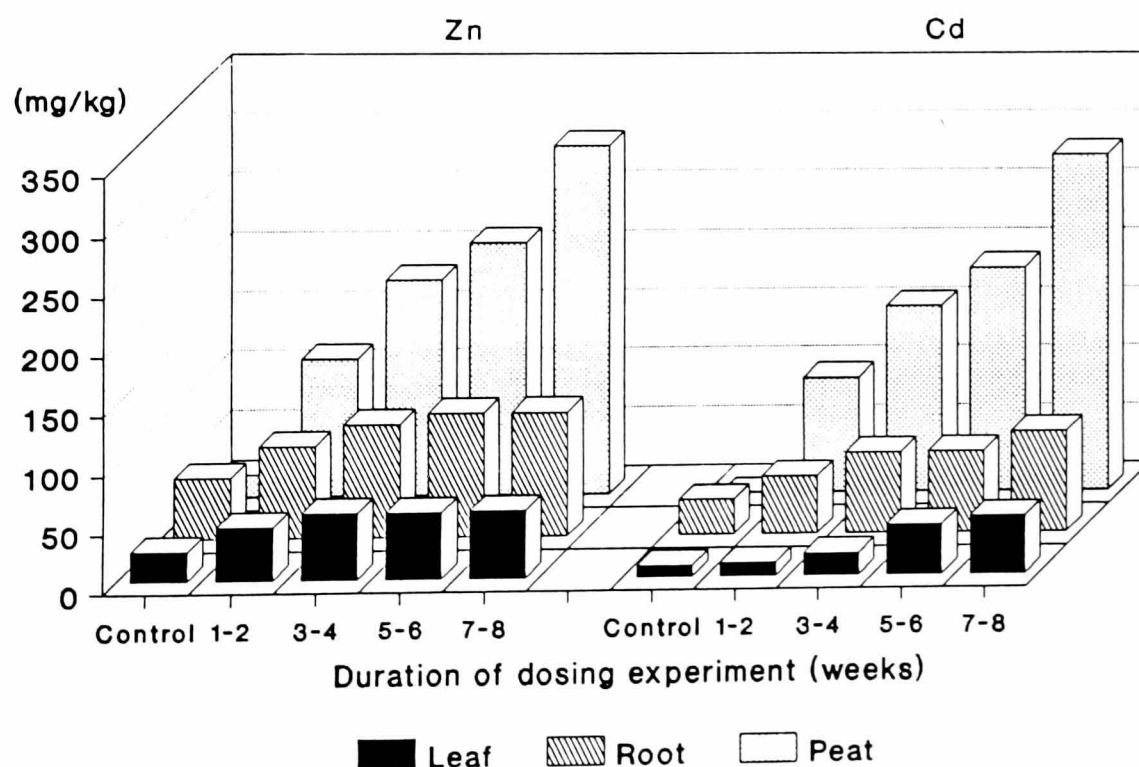
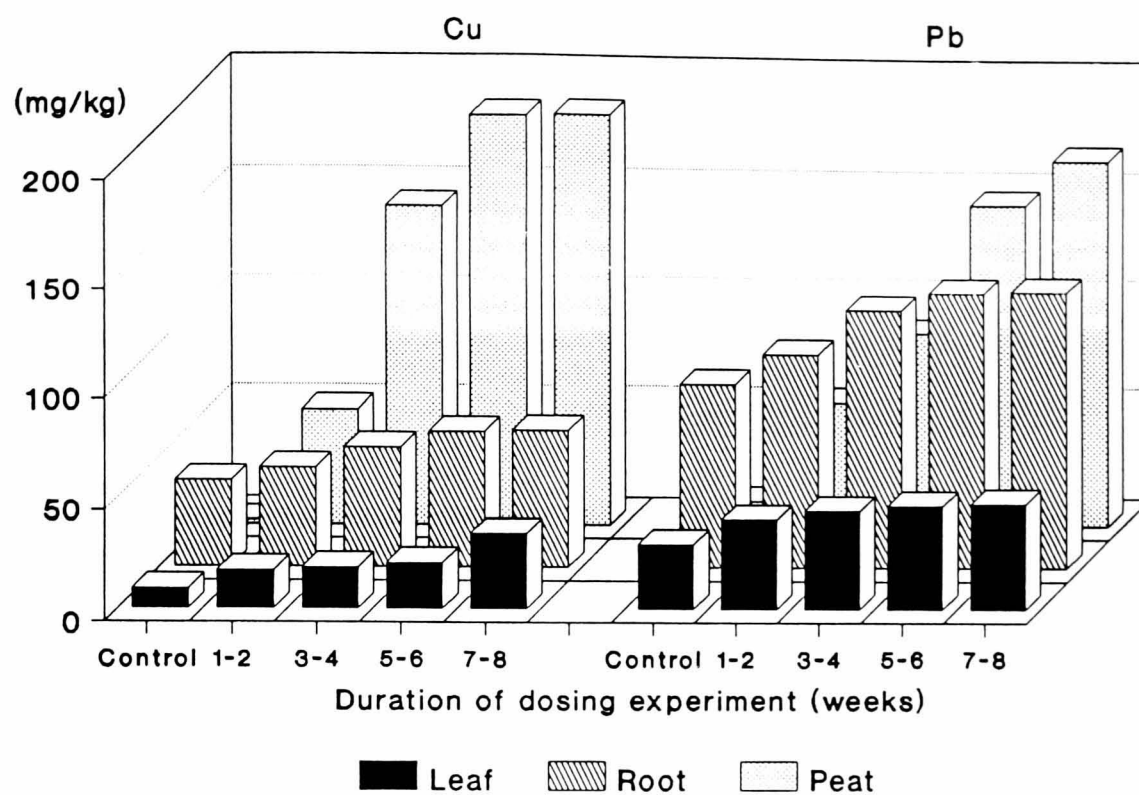
* values would be in mg if based on extraction from 1kg dry weight root tissue

4.2.2.2 Long term dosing experiments with Juncus effusus

Greenhouse-based, long term dosing experiments with Juncus were also carried out over an eight week period. During this test, peat and plants were dosed with (10 ppm) mixed metal solution. Surface peat metal levels demonstrated substantially enhanced metal levels over the eight week experimental period with increases observed rising from 0 to 314.0, 2 to 198, 11.2 to 203, and 19.2 to 323 mg/kg for Cd, Cu, Pb and Zn respectively (Fig.4.5). All metal levels in Juncus root tissues increased consistently if only at lower rates over the 8 weeks experimental period, but compared to the metal elevation patterns observed for the sediment, the root tissues patterns are more consistent. The metal elevation of plant leaf tissues showed a reduced affinity but nevertheless similar patterns of small consistent increases can be observed (Fig.4.5).

In comparison to the uptake of Cu and Pb, the roots show a higher uptake ability for Zn and Cd. After 8 weeks, metal concentrations increased by a factor of 2.8, 2.0, 1.6 and 1.5 for Cd, Zn, Cu and Pb respectively. This would correspond to Cd and Zn established equilibria noted in urban waters as being predominantly soluble bioavailable metal ions or as weak complexes (Revitt and Morrison, 1987).

The observed peat, Juncus root and leaf metal uptake data over the eight week time period of the dosing experiment have also been analysed by statistical regression methods. The resulting equations are shown in Fig.4.6. Juncus leaf Cu and Cd uptake patterns are exponential whereas Pb and Zn uptake patterns are linear. The root uptake trends for all metals are linear.



Dosing concentration 10 ppm
Dosing time 3/10/1989 - 29/11/1989

Fig.4.5 Temporal increases in peat and Juncus tissue metal concentrations during 8 week dosing experiments

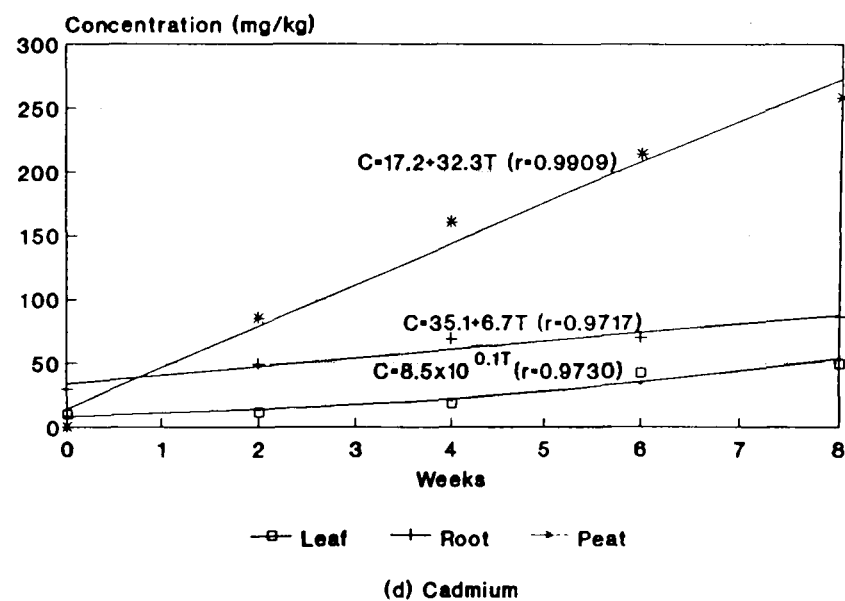
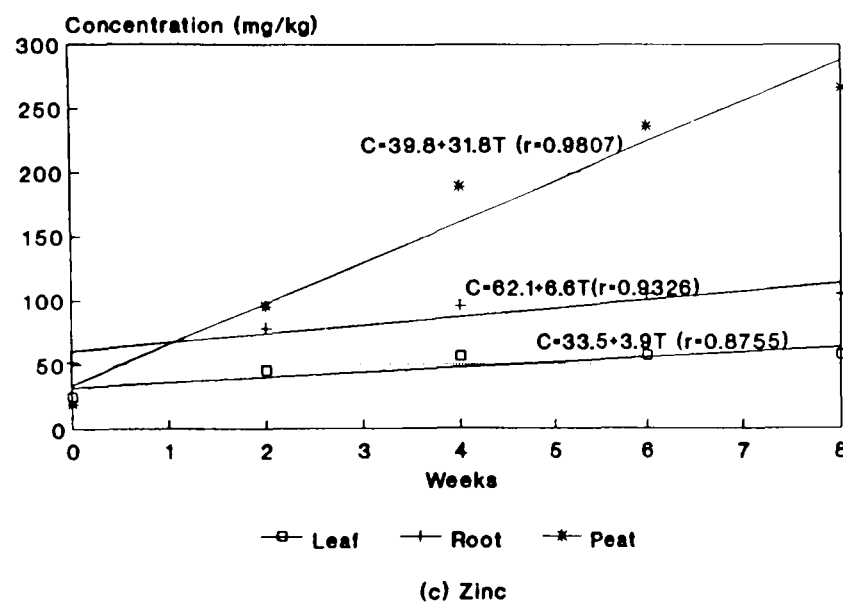
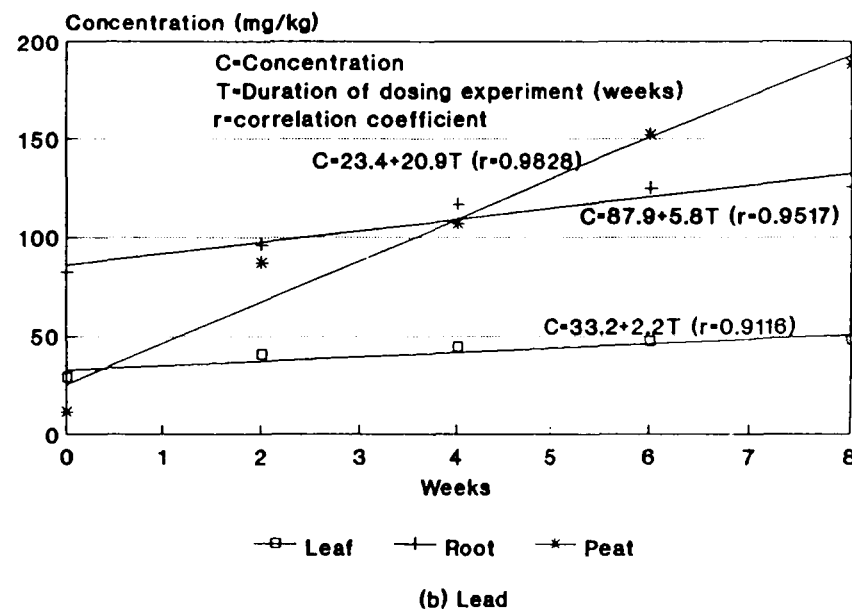
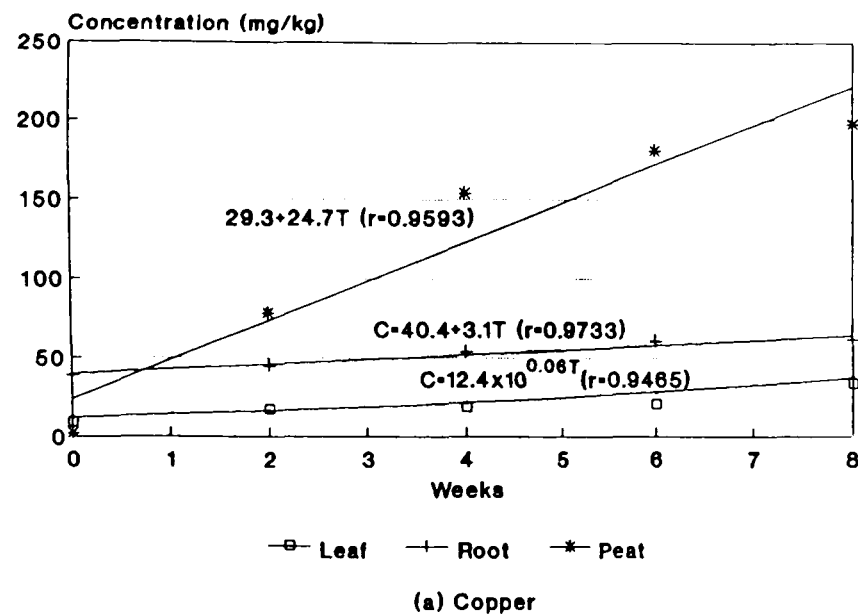


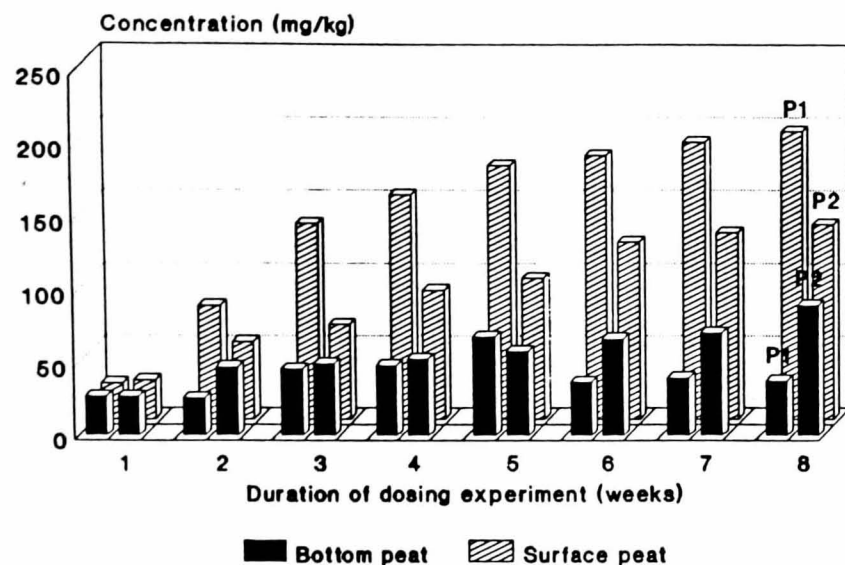
Fig.4.6 Regression equations and graphs of metal uptake by peat and Juncus tissues during 8 week dosing experiments

After the 8 weeks dosing experiment, the root to leaf ratios of Juncus plants decreased from 4.3 to 1.8, 3.2 to 1.8, 2.1 to 1.8, and 2.8 to 2.6 for Cu, Cd, Zn and Pb respectively. This suggests that the metals taken up by the roots are being transferred to the leaf efficiently and in the order of Cu>Cd>Zn>Pb. Previous studies have demonstrated the translocation of Cd from roots to above ground parts in water hyacinth (Blake et al., 1987).

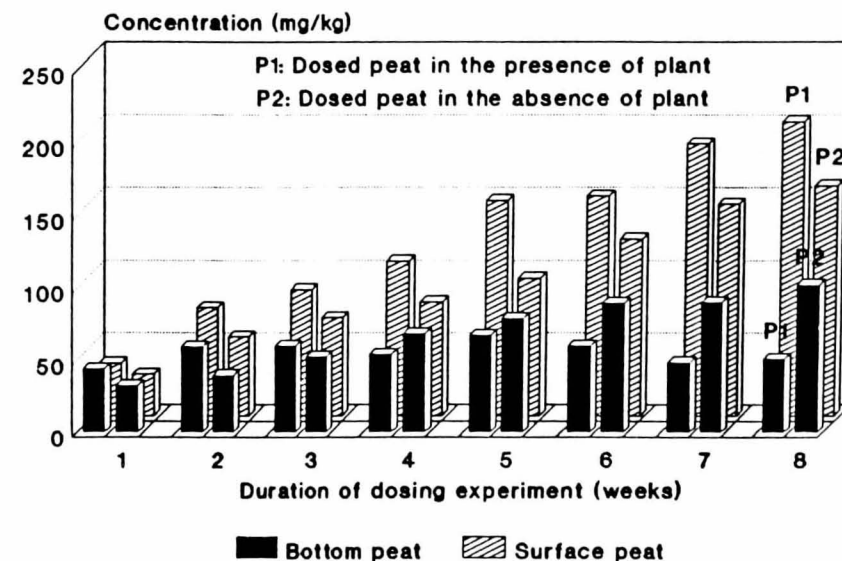
4.2.2.3 Long term dosing experiment with peat

An additional 8 week dosing experiment was also carried out with peat to examine the metal uptake of the substrate material in the absence of plants. The results of this experiment are compared with the peat metal uptake data obtained from the long term dosing experiment with Typha latifolia (Fig. 4.7).

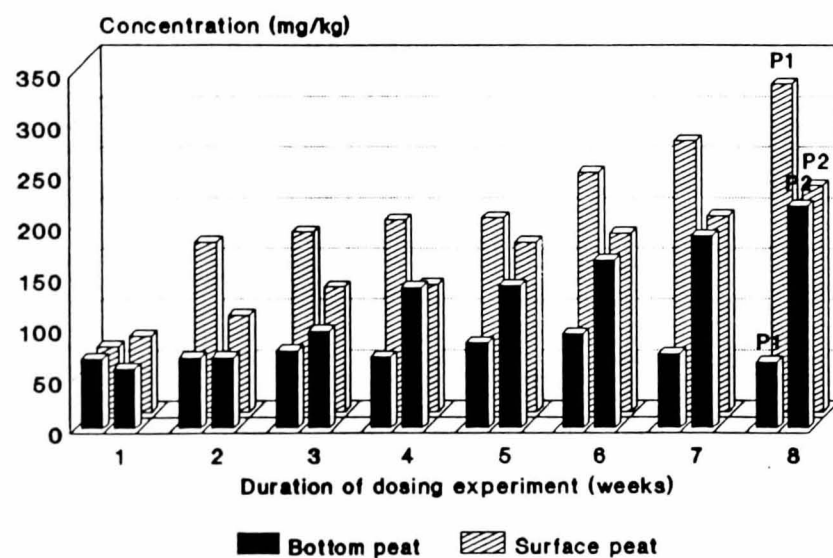
Surface peat concentrations are increased progressively for all metals (Fig.4.7) but are all lower than the surface peat metal levels recorded in the case of the dosing experiment conducted in the presence of Typha. The bottom peat concentrations of all metals also increased consistently with Zn and Cd levels equilibrating throughout the peat profile at end of the experiment. Basal peat Cu and Pb levels however remain lower than the surface levels.



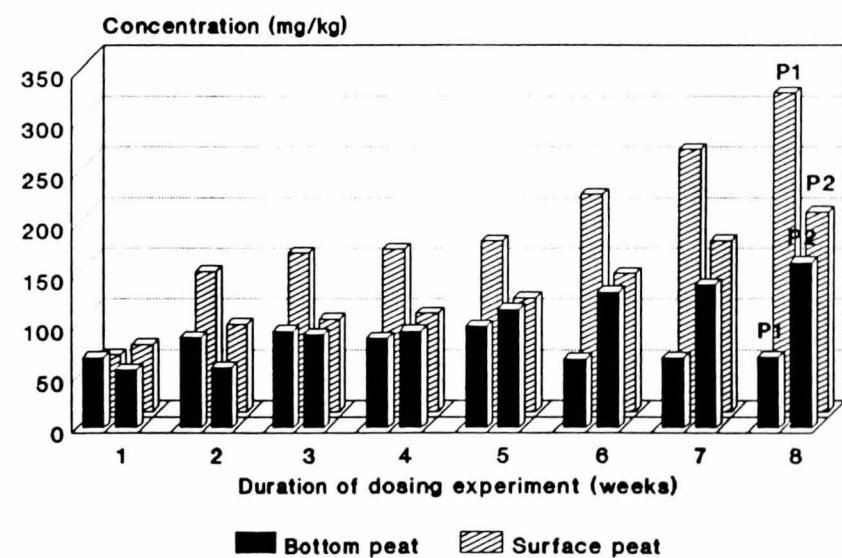
(a) Copper



(b) Lead



(c) Zinc



(d) Cadmium

Dosing concentration 10 ppm

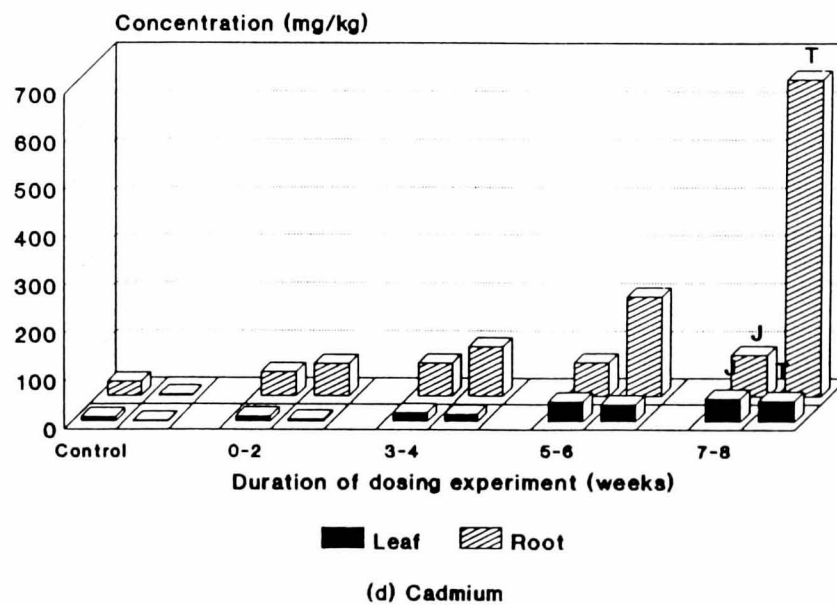
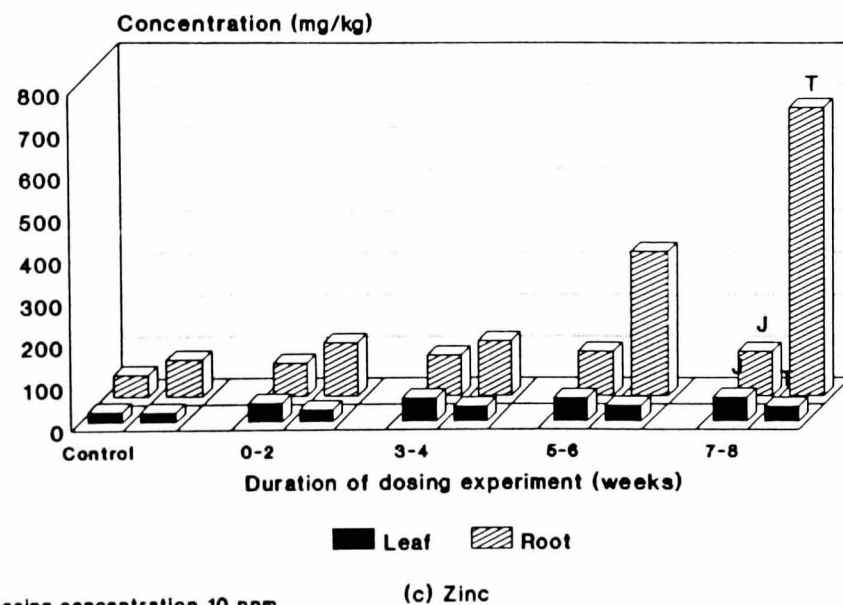
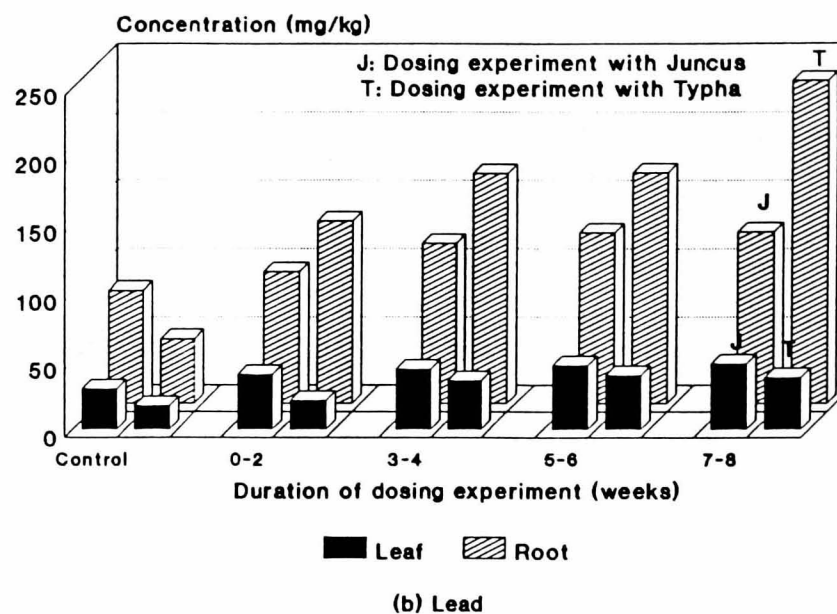
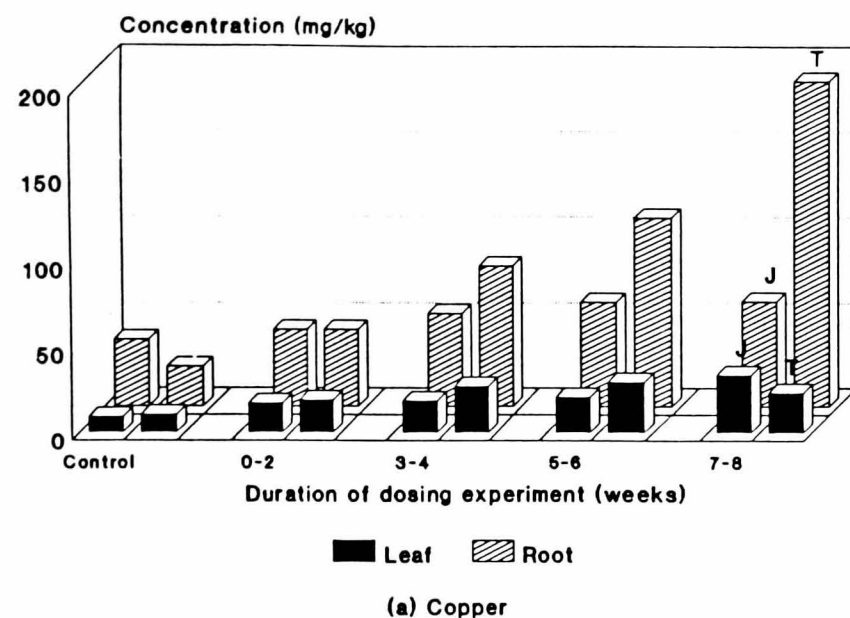
Fig.4.7 Metal levels in surface and bottom peat in the presence or absence of Typha during 8 week dosing experiments

Comparison of the bottom peat metal levels showed that after 5 weeks, the metal levels of bottom peat without plant continued to increase, whereas peat with Typha levels decreased. Metal and associated water uptake by Typha roots is clearly resulting in the relatively lower observed bottom peat concentrations, and the decrease in levels after 5 weeks is possibly a consequence of an increase in ambient temperatures and also the evapotranspiration rate.

4.2.2.4 Comparison of dosing experiments with Typha latifolia and Juncus effusus

The results of the 8 week dosing experiments with Typha were compared with those obtained with Juncus (Fig. 4.8).

All metal levels in Typha root tissues show a constant uptake with Zn and Cd levels up to 689 and 662.5 mg/kg respectively. The metal root level ratios of 8 week dosed to controlled plant are 8.3, 5.1, 7.8 and 200.8 for Cu, Pb, Zn and Cd respectively (Table 4.2). In comparison to Typha, metal levels in the dosed Juncus root are relatively stable. The metal root level ratios of dosed to controlled Juncus at any dosing duration are lower than those for Typha. Although the experiments were not conducted simultaneously, the data indicate that Typha roots have a higher ability for metal uptake.



Dosing concentration 10 ppm

Fig.4.8 Metal levels in Typha latifolia and Juncus effusus during 8 week dosing experiments

Even though all metal levels in Juncus leaf tissues are higher than the levels recorded in Typha leaf tissues after 8 weeks, the metal ratios of dosed to controlled plant for Cu (except 7-8 weeks dosed/control), Pb and Cd are lower than the ratios of Typha. Juncus Zn level ratios of dosed to controlled plant are all higher than the ratios of Typha. This suggests that the translocation of Zn from root to leaf of Juncus is faster than for Typha, whereas Typha shows a higher affinity for Cd, Cu and Pb.

Table 4.4: Comparison of metal root and leaf ratios
between controlled and dosed plant

		root				
		Control	1-2	3-4	5-6	7-8 (weeks)
Cu	<u>Typha</u>	1 (23.0)	1.9	3.6	4.8	8.3
	<u>Juncus</u>	1 (39.0)	1.1	1.4	1.5	1.6
Pb	<u>Typha</u>	1 (47.2)	2.8	3.5	3.6	5.1
	<u>Juncus</u>	1 (82.5)	1.2	1.4	1.5	1.5
Zn	<u>Typha</u>	1 (87.6)	1.5	1.5	3.9	7.8
	<u>Juncus</u>	1 (52.0)	1.5	1.8	2.0	2.0
Cd	<u>Typha</u>	1 (3.3)	20.5	31.2	63.3	200.8
	<u>Juncus</u>	1 (30.0)	1.6	2.3	2.3	2.8
		leaf				
		Control	1-2	3-4	5-6	7-8 (weeks)
Cu	<u>Typha</u>	1 (10.4)	1.8	2.6	2.9	2.3
	<u>Juncus</u>	1 (9.0)	1.8	2.1	2.3	3.8
Pb	<u>Typha</u>	1 (17.2)	1.3	2.1	2.3	2.2
	<u>Juncus</u>	1 (29.0)	1.4	1.5	1.6	1.7
Zn	<u>Typha</u>	1 (23.2)	1.2	1.6	1.6	1.6
	<u>Juncus</u>	1 (24.7)	1.8	2.3	2.3	2.3
Cd	<u>Typha</u>	1 (2.4)	2.1	6.5	15.2	18.3
	<u>Juncus</u>	1 (9.5)	1.2	1.9	4.5	5.2

4.3 Conclusions

The major findings of this section of the research can be summarised as follows:-

a) The short term greenhouse-based metal uptake experiments show that metal levels in dosed peat, Typha root, rhizome and leaf increase after 10 days and when the dosing water concentrations were increased, the concentration differences between peat, root, rhizome and leaf also increased in parallel. Dosed peat and Typha roots show a greater uptake ability for Cd and Cu than for Zn and Pb.

b) Long term greenhouse-based metal uptake experiments with Typha showed a greater enhancement of metal levels by surface peat over the eight week experimental period, whereas bottom peat metal levels remain relatively stable. The surface peat metal levels are up to 4.8 higher than basal metal levels. The surface sediment increases are most closely paralleled by metal uptake patterns in the Typha root, with Zn and Cd both exhibiting exponential increases with time and final values approaching 700 mg/kg. The uptake ability is in the order of Cd>Cu>Zn>Pb. The root to rhizome translocation is most efficient for Cu followed by Pb, with Cd and Zn showing a preferential metal accumulation within the root zone.

c) Long term dosing experiment with Juncus effusus also shows an enhancement of peat metal uptake, but metal elevation patterns of root tissues are more consistent over time. In comparison to Cu and Pb, roots

translocation efficiency is in the order of Cu>Cd>Zn>Pb.

d) The long term dosing experiments with peat show that the surface peat concentrations of all metals increased constantly but are all lower than the surface peat metal levels recorded in the dosing experiments conducted in the presence of plants over a 1 week period. After 5 weeks, the metal levels of basal peat in the absence of the plant continue to increase, whereas the metal levels of bottom peat with plants decrease.

e) The comparison of dosing experiments with Typha latifolia and Juncus effusus which were not carried out simultaneously suggest that the former plants have a higher ability for metal uptake. The results also show that the metal translocation from root to leaf is more efficient in Typha for Cd, Cu and Pb whereas Juncus is more efficient for Zn translocation.

Chapter 5: Field Studies of Metal Uptake by Typha latifolia

5.1 Introduction

Man's activity has caused a drastic changes in metal fluxes within the biosphere (Grahn and Hakanson, 1987). In recent years, the fluxes of many trace metals from terrestrial and atmospheric sources to the aquatic environment have substantially increased (Forstner & Wittmann, 1981). When a trace metal is added to a natural water system, it partitions amongst various compartments. As a result of complex physical, chemical and biological processes, a major fraction of the trace metal introduced into the aquatic environment eventually becomes associated with the bottom sediment (Tessier and Campbell, 1987). Metals present in the water and sediment can then be taken up by aquatic macrophytes. Emergent plants obtain all their minerals from the sediment (Haslam, 1987). Studies have also shown seasonal variations to occur in metal uptake of the emergent species such as Phragmites sp (Larsen and Schierup, 1981; Mortimer, 1985).

The aim of this part of the study is to determine metal uptake under field conditions by sediment and Typha latifolia plants.

Typha latifolia plants and their associated sediment and surrounding waters within a rural pond (site 1) and two urbanized wetlands (sites 2 and 3) receiving surface discharges were analysed. Seasonal variations of Cd, Cu, Pb and Cd in sediment, water, and Typha root, rhizome and leaf tissues were determined. The metal levels in sediment were then compared with those recorded in plant root,

rhizome and leaf tissues.

5.2 Discussion of the Results

5.2.1 Heavy Metal Levels in Wetland Water Samples

Ranges and average metal concentrations monitored over a 14 month field study for the water phase are shown in Fig.5.1. The average Cu, Pb, Zn and Cd levels at the two urban sites (sites 2 and 3) are higher than those recorded at the rural site (site 1) with site 3 being the most polluted. The concentration differences of all metals between site 3 and site 1 are statistically significant (Table 5.1). The concentration differences of Cu and Cd between site 2 and site 1 are significant, whereas the differences of Pb and Zn are not. The ratios of mean metal levels for the sites (3:2:1) are: Cu, 4.8:2.9:1, Pb, 2.0:1.2:1, Zn, 2.0:1.3:1 and Cd, 2.2:1.4:1.

The water metal levels show a large seasonal variability at the two urban sites whereas the metal levels, with the exception of Zn, in water at site 1 are relatively stable (Table 5.2).

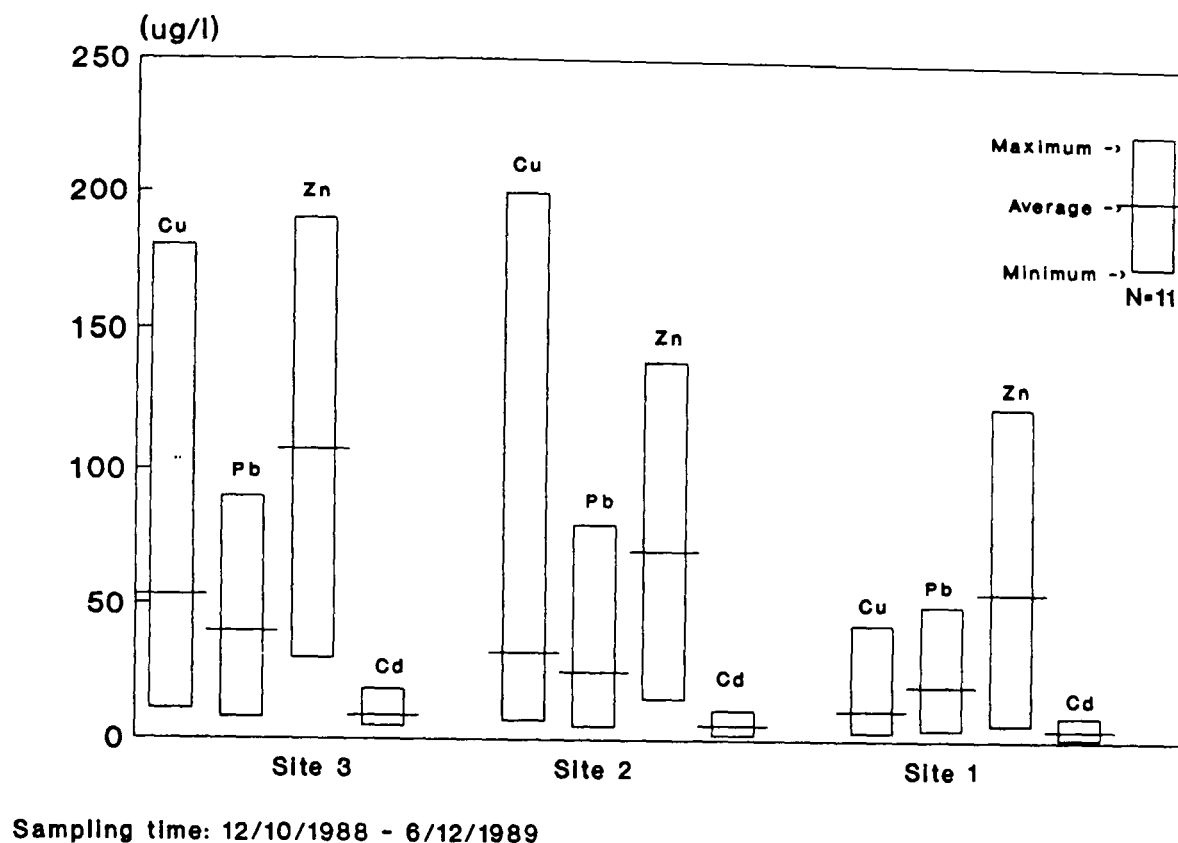


Fig.5.1 Average and ranges of heavy metal concentration in wetland water samples

Table 5.1: T values of water samples using Students 't' test

	$t_{0.05,11}$ (site 1 and site 2)	$t_{0.05,11}$ (site 1 and site 3)
Cu	1.865 (>1.796)	3.928 (>1.796)
Pb	0.865 (<1.796)	1.914 (>1.796)
Zn	1.281 (<1.796)	2.643 (>1.796)
Cd	3.324 (>1.796)	1.904 (>1.796)

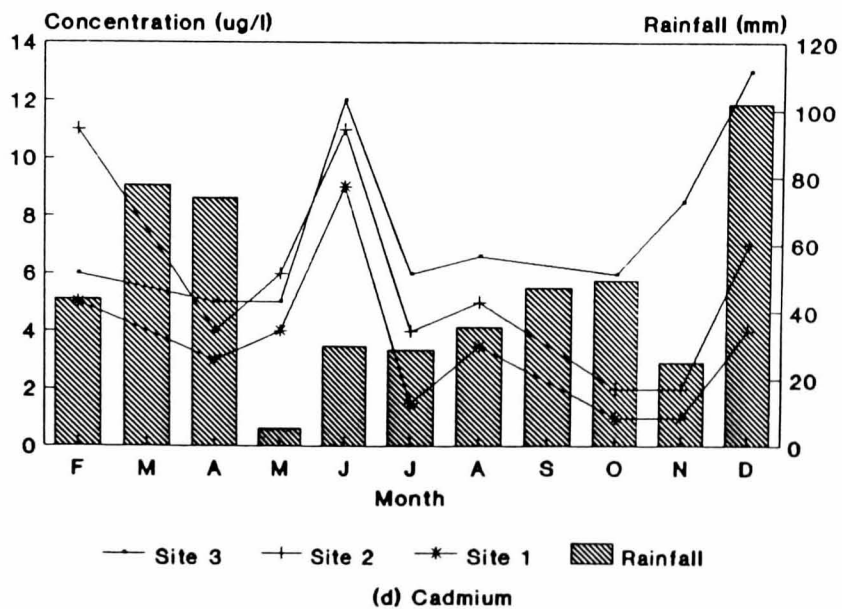
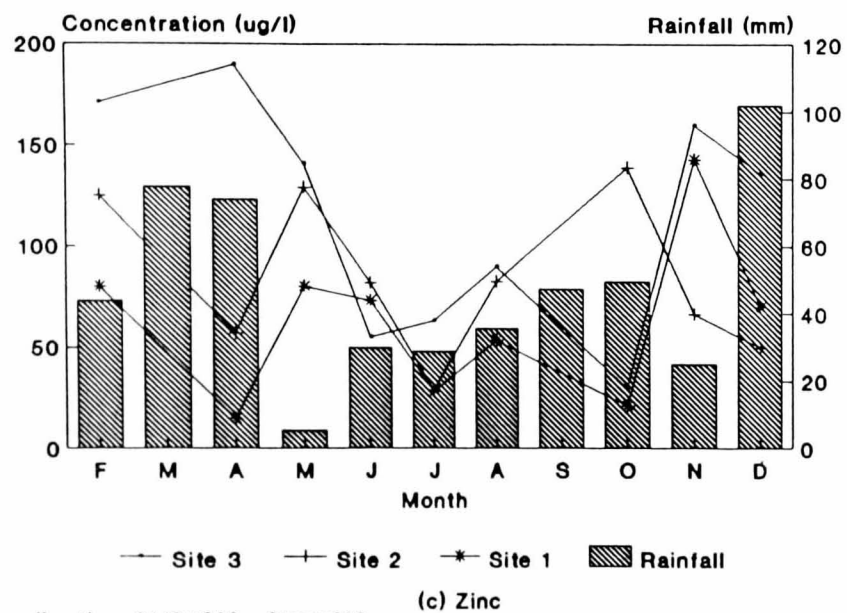
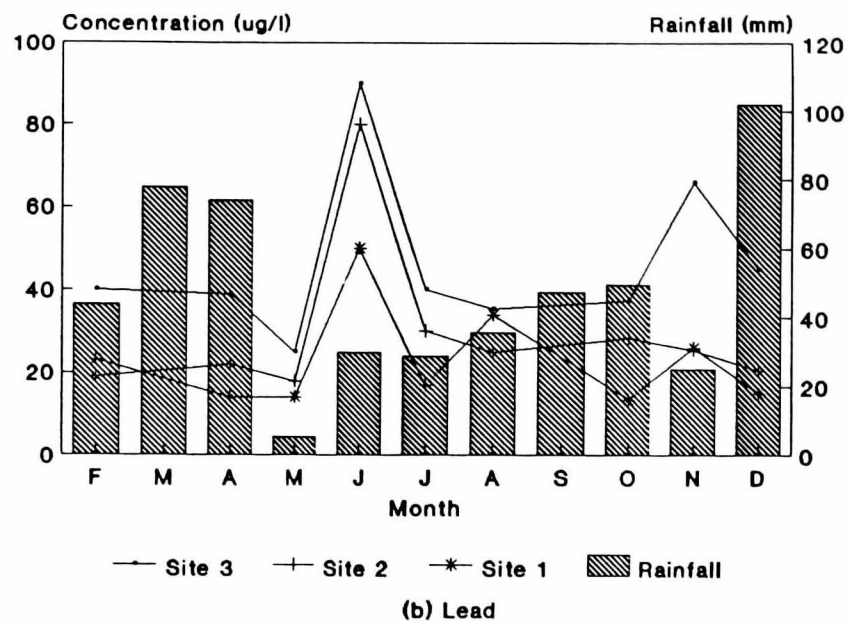
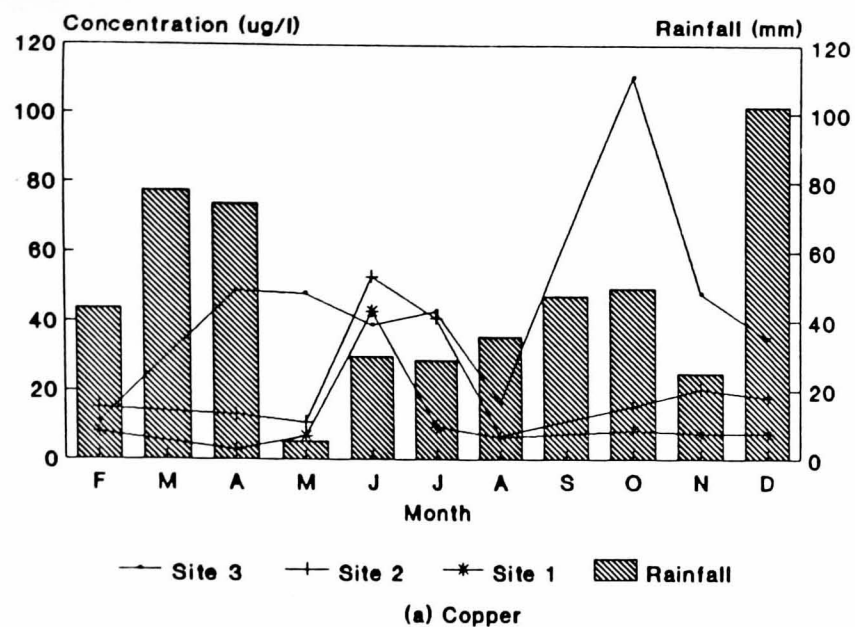
when $n=11$, $P<0.05$, $t_{0.05,11}>1.796$, the difference is significant

Table 5.2: Average and standard deviation
of metal levels in water samples (ug/l)

	Site 1	Site 2	Site 3
Cu	17.0 ± 21.3	39.4 ± 55.2	59.6 ± 46.0
Pb	23.8 ± 22.3	27.4 ± 18.8	36.2 ± 15.7
Zn	63.0 ± 42.1	77.5 ± 40.4	136.6 ± 107.1
Cd	5.3 ± 4.5	6.0 ± 3.1	8.9 ± 4.2

The overall temporal changes in water metal levels at the three sites and corresponding rainfall levels are shown in Fig.5.2 a-d. Copper, Pb and Cd levels at all sites show elevations in late spring/early summer which may be a consequence of storm events; this is most noticeable in the case of Pb and Cd. Copper levels at site 3 reach the highest value 116 ug/l in October following earlier lower summer peaks also associated with storm events. There is no clear trend in Zn levels which can be discerned at any of the sites although there is a tendency of a summer 'depression' in the observed values. The average Zn and Pb levels (107.3 and 39.8 ug/l) of the most polluted site (site 3) are similar to those recorded by Prahalad and Seenayya (1989) in a polluted lake.

In aquatic environments, water is the primary contaminated medium and an inherently high degree of variation in metal concentration naturally arises. Metals that do not remain soluble in the water phase become accumulated within the bottom sediment through sedimentation, thus the metal levels found in the sediment are affected by the concentration of metal levels in the ambient water.

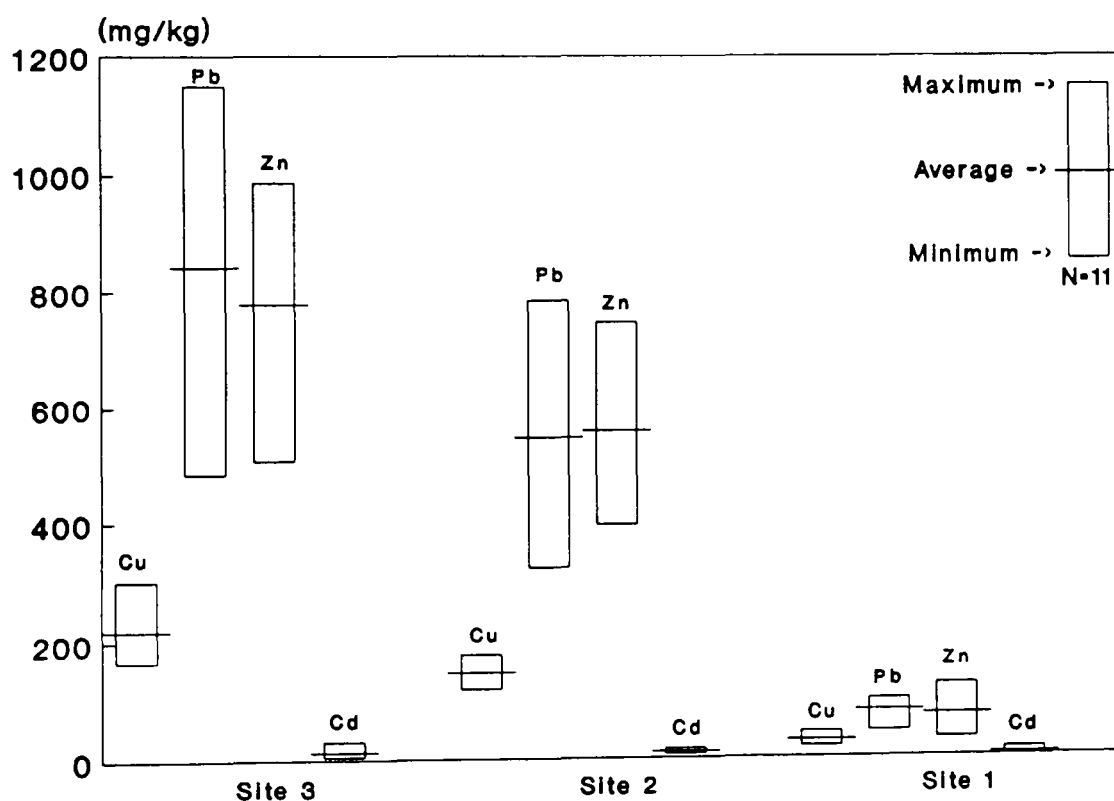


Sampling time: 24/2/1989 - 6/12/1989

Fig. 5.2 Temporal variation of heavy metal levels in wetland water samples

5.2.2 Heavy Metal Levels in Wetland Sediments

Average metal concentrations together with the ranges monitored during the 14 month field study for sediment samples are shown in Fig.5.3. Copper, Pb, Zn and Cd levels exhibit the expected elevations at the two urbanised sites (sites 2 and 3) compared to the rural site (site 1). The differences in all metal levels between both urban sites and the rural site are statistically significant (Table 5.3). The ratios of mean metal levels for the sampling sites (3:2:1) are: Cu, 7.8:5.2:1, Pb, 10.8:7.0:1, Zn, 11.0:7.9:1 and Cd, 3.0:2.6:1.



Sampling time: 12/10/1988 - 6/12/1989

Fig.5.3 Average and ranges of heavy metal concentration in wetland sediment

Table 5.3: T values of sediment samples
using Students 't' test

	$t_{0.05,11}$ (site 1 and site 2)	$t_{0.05,11}$ (site 1 and site 3)
Cu	31.788 (>1.796)	14.037 (>1.796)
Pb	10.559 (>1.796)	10.947 (>1.796)
Zn	12.118 (>1.796)	18.171 (>1.796)
Cd	5.953 (>1.796)	4.667 (>1.796)

when $n=11$, $P<0.05$, $t_{0.05,11}>1.796$, the difference is significant

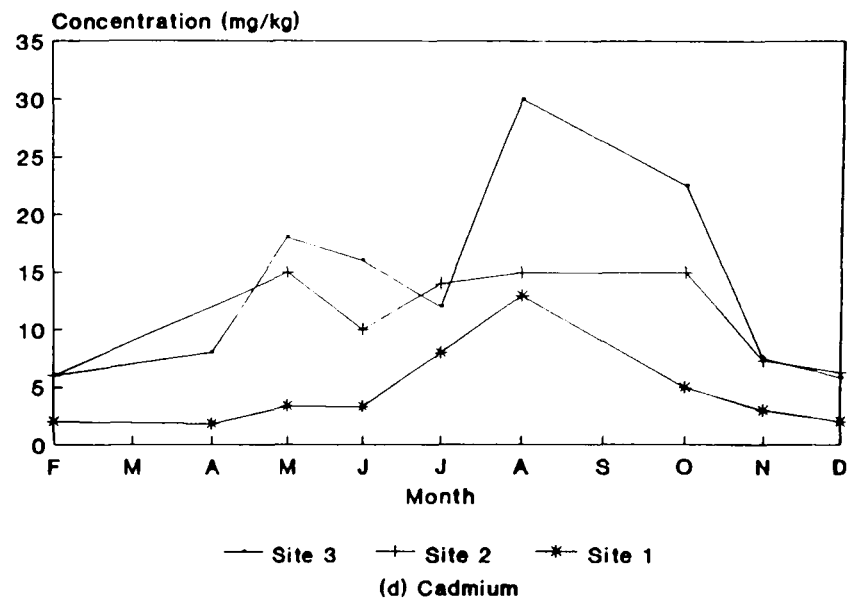
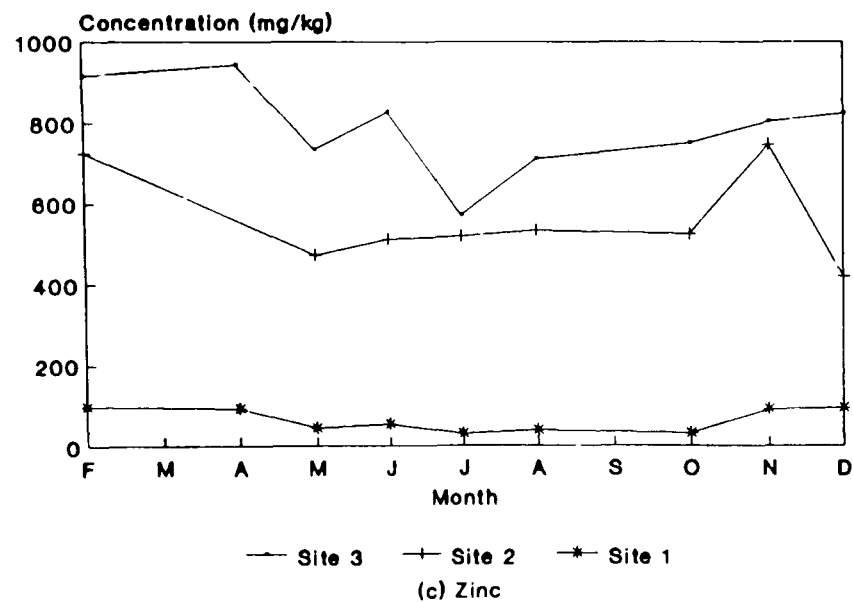
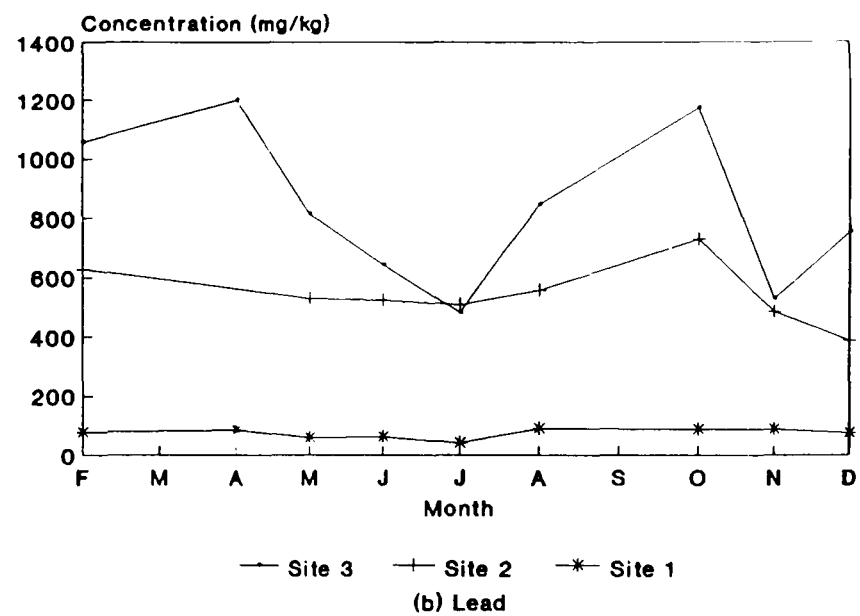
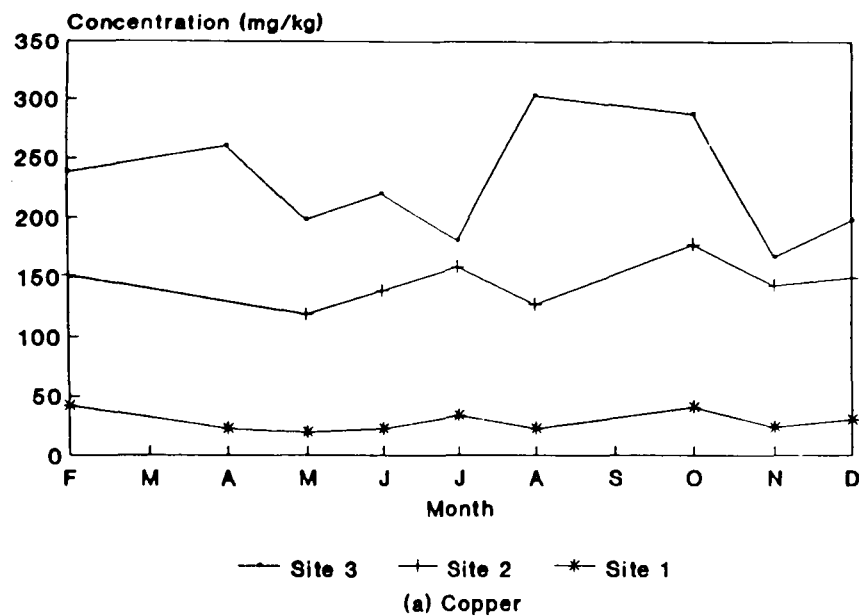
All sediment levels show a high variability at the two urban sites with the greatest variation being observed at site 3. Metal levels in the rural site sediment are very much more stable (Table 5.4). The average Cu and Zn levels of the two urban sites are similar to those recorded by Oliver (1973), Aulio (1980) and Prahalad and Seenayya (1989) in polluted rivers and lakes. These results are between McNaughton et al. (5000 mg/kg for Zn) (1974) and Taylor and Crowder's (3738 mg/kg for Cu, 343 mg/kg for Zn) (1983) results. The average Pb levels of two urban sites 841.2 and 545.4 mg/kg are higher than those recorded by Oliver (390 mg/kg, 1973), McNaughton et al. (435 mg/kg, 1974) and Prahalad and Seenayya (59.7 ± 20.0 mg/kg, 1989). The sediment Cd levels at the two urban sites are much lower than those recorded by McNaughton et al. (73 mg/kg) (1974).

Table 5.4: Average and standard deviation
of metal levels in sediment samples (mg/kg)

	Site 1	Site 2	Site 3
Cu	28.2 ± 7.9	119.9 ± 58.4	219.8 ± 44.5
Pb	77.7 ± 15.6	496.2 ± 202.0	841.2 ± 228.8
Zn	70.7 ± 30.6	506.0 ± 197.1	778.9 ± 139.6
Cd	4.2 ± 3.3	9.7 ± 4.8	12.5 ± 7.8

There is field evidence for contaminated sediment flushing occurring to the receiving basin (site 3) during late summer/early autumn following storm events, especially in the case of Cu, Pb, and Cd (Fig.5.4 a-d). This follows the early summer peak levels of Cu, Pb and Cd observed in the water phase. These events appear to have entrained and incorporated toxic materials within the mud substrate of the receiving basin. Zn sediment levels stay relatively stable during this period at all sites, corresponding to the trends noted in the water levels.

Sediments play a crucial role in both ambient water quality and in plant metal uptake. They act as a sink to remove metal pollutants from the water column and may release metal contaminants to surface waters. The amount of toxic heavy metals present in contaminated sediment is considered to be the amount potentially available to the plants. Emergent plants obtain all their minerals from the sediment (Haslam, 1987), therefore a proper assessment of metal levels in the sediment is essential when considering potential metal uptake by aquatic plants.



Sampling time: 24/2/1989 - 6/12/1989

Fig.5.4 Temporal variations of heavy metal levels
in wetland sediment

5.2.3 Heavy Metal Levels in Typha Root Tissues

The highest Typha metal levels are recorded in the root tissue. Average metal concentrations together with ranges monitored during the 14 month field study for Typha root tissues samples are shown in Fig.5.5. Corresponding to the sediment metal levels, Cu, Pb, Zn and Cd levels exhibit the expected elevations at the two urbanised sites (sites 2 and 3) compared to the rural site (site 1). The differences of all metal levels between urban sites and rural site are statistically significant (Table 5.5). The metal ratios for the sampling sites (3:2:1) are: Cu, 1.9:1.4:1, Pb, 2.0:2.0:1, Zn, 1.9:1.9:1 and Cd, 1.8:1.5:1.

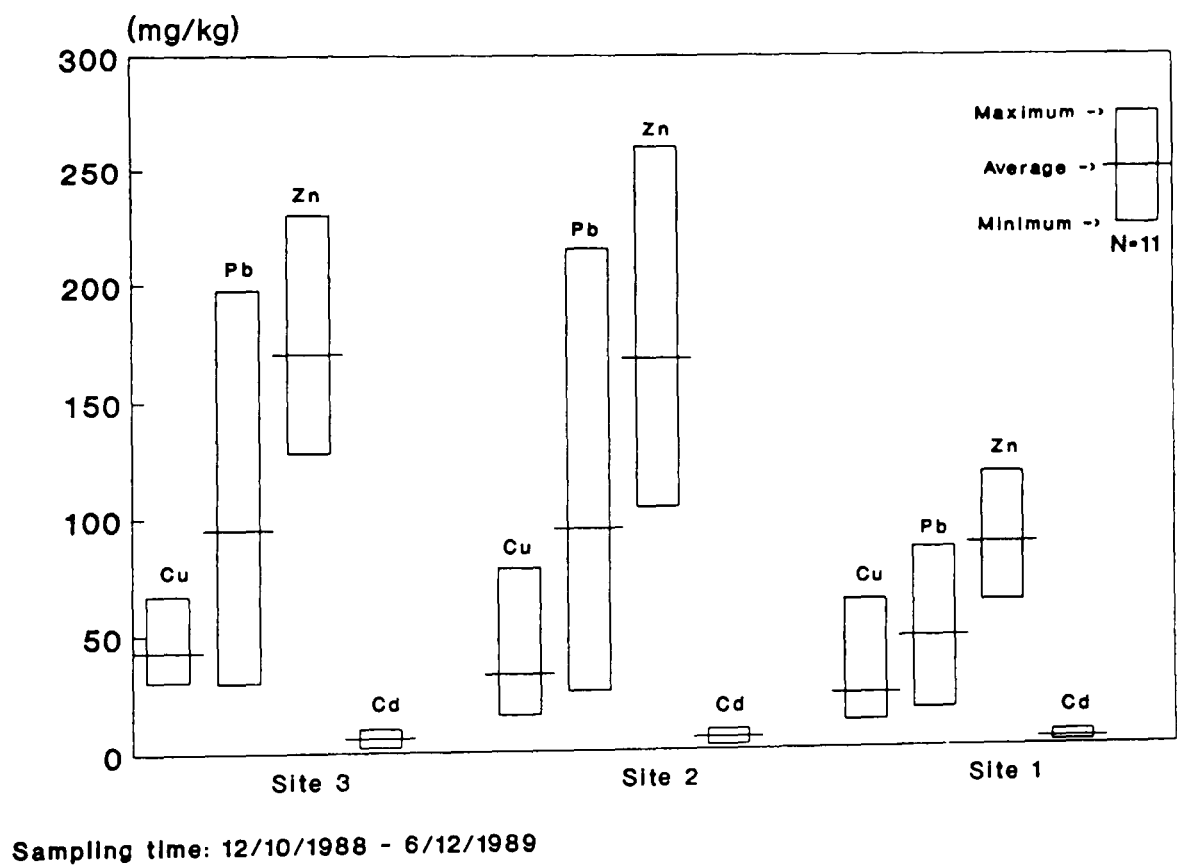


Fig 5.5 Average and ranges of heavy metal concentration in Typha root tissues

Table 5.5: T values of Typha root samples
using Students 't' test

	$t_{0.05,11}$ (site 1 and site 2)	$t_{0.05,11}$ (site 1 and site 3)
Cu	4.386 (>1.796)	4.865 (>1.796)
Pb	3.382 (>1.796)	3.685 (>1.796)
Zn	4.795 (>1.796)	10.371 (>1.796)
Cd	4.978 (>1.796)	6.377 (>1.796)

when $n=11$, $P<0.05$, $t_{0.05,11}>1.796$, the difference is significant

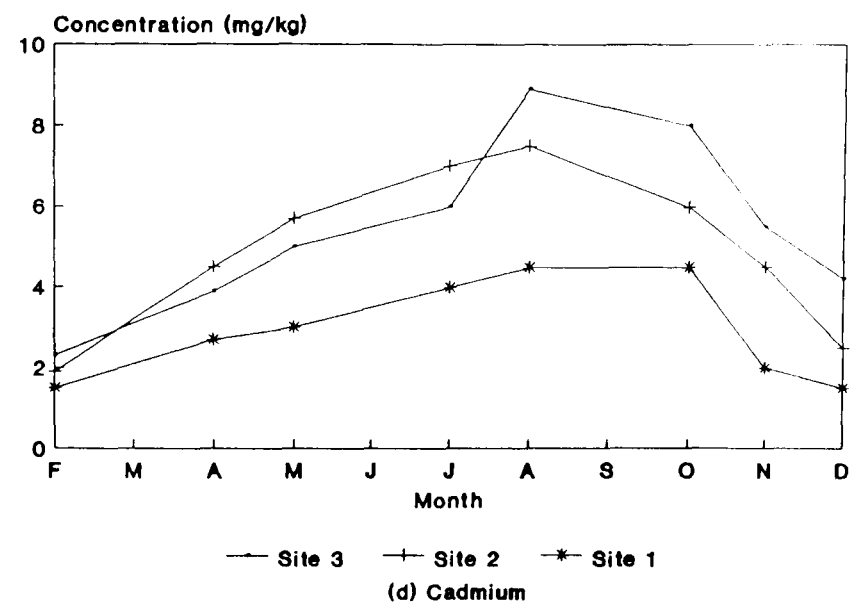
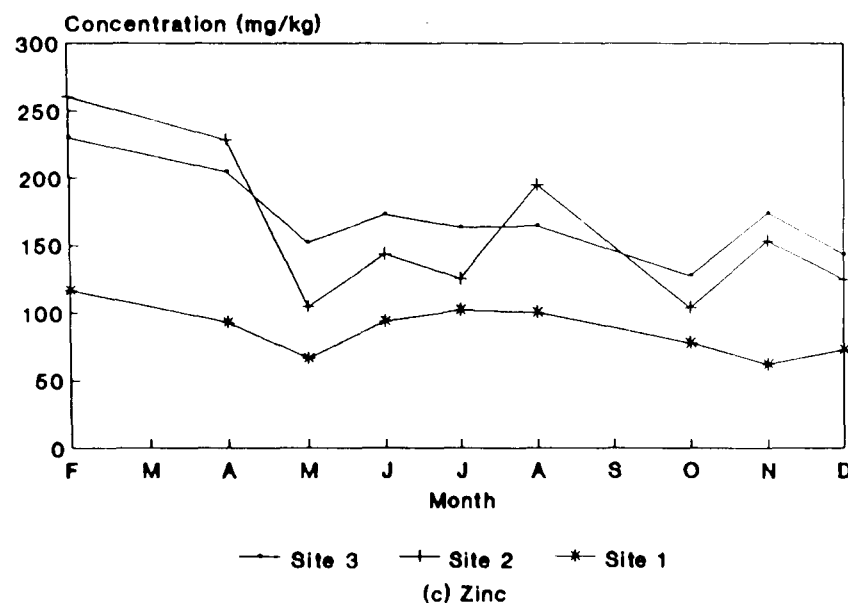
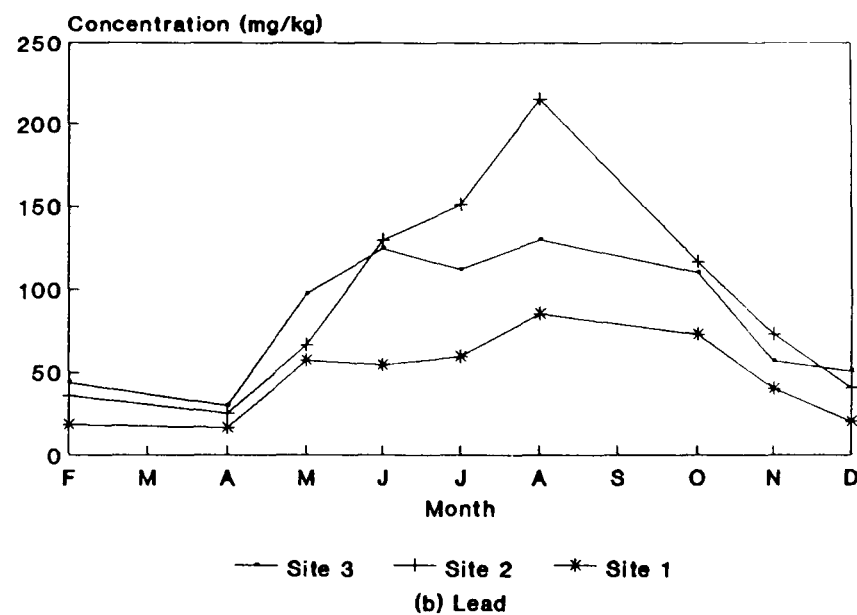
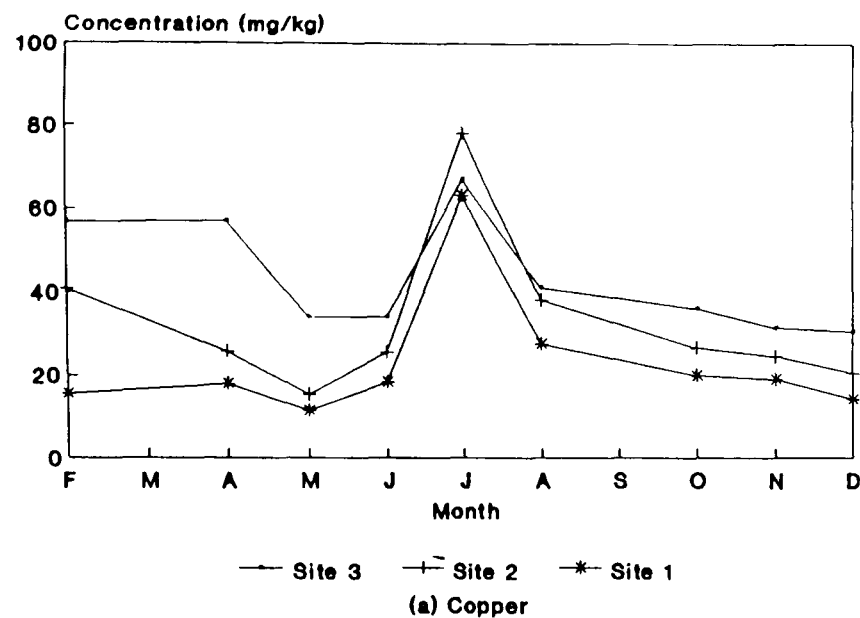
The Cu, Pb, Zn and Cd levels in root tissues show considerable temporal variability (Table 5.6), with peak levels of Cu occurring during the growth period which may be associated with an expected increase in demand for plant micronutrients (Fig.5.6 a). The maximum Pb and Cd root levels occur during August to October following bioaccumulation (Fig.5.6 b,d), As in the case of sediment and water, Zn concentrations in the plant root tissues at all three sites do not demonstrate any distinct seasonal variations (Fig.5.6 c) as do the other metals. However, Zn concentrations fall in May corresponding with translocation to rhizome and leaf during growth. The average root Pb levels at the two urban sites are similar to those obtained from Potamogeton robbinsii root by Reimer (1989).

Table 5.6: Average and standard deviation
of metal levels in Typha root samples (mg/kg)

	Site 1	Site 2	Site 3
Cu	23.0 ± 14.7	32.7 ± 17.7	39.7 ± 17.7
Pb	47.3 ± 23.5	94.5 ± 59.4	85.6 ± 56.1
Zn	87.5 ± 17.6	160.1 ± 52.6	153.5 ± 58.2
Cd	3.3 ± 1.5	5.0 ± 2.2	5.4 ± 2.9

Root Cu and Zn levels at the two urban sites are within the range of the results (13 - 265 mg/kg for Cu and 24 - 572 mg/kg for Zn) of a study of metal accumulation of Typha latifolia near smelters (Taylor, 1983).

Emergent plants like Typha latifolia absorb relatively few nutrients or metals though their thick rhizomes or aerial parts (Haslam, 1987). Root functions therefore play a crucial role in metal uptake for emergent plant species.



Sampling time 24/2/1989 - 6/12/1989

Fig.5.6 Temporal variations of Heavy metal levels
in Typha root tissues

5.2.4 Heavy Metal Levels in Typha Rhizome Tissues

Average metal concentrations together with ranges monitored during the 14 month field study of Typha rhizome samples are shown in Fig. 5.7. The average metal levels in rhizome tissues are less than the metal levels in root but greater than those in leaf tissue. Copper, Pb, Zn and Cd levels at the two urbanised sites (site 2 and 3) are consistently higher than those at the rural site (site 1). The differences of all metal levels between urban sites and the rural site are statistically significant (Table 5.7). The ratios of the average metal levels in Typha rhizome tissues for the sites (3:2:1) are: Cu, 1.8:1.5:1, Pb, 1.4:1.2:1, Zn, 1.8:1.4:1 and Cd, 1.5:1.3:1.

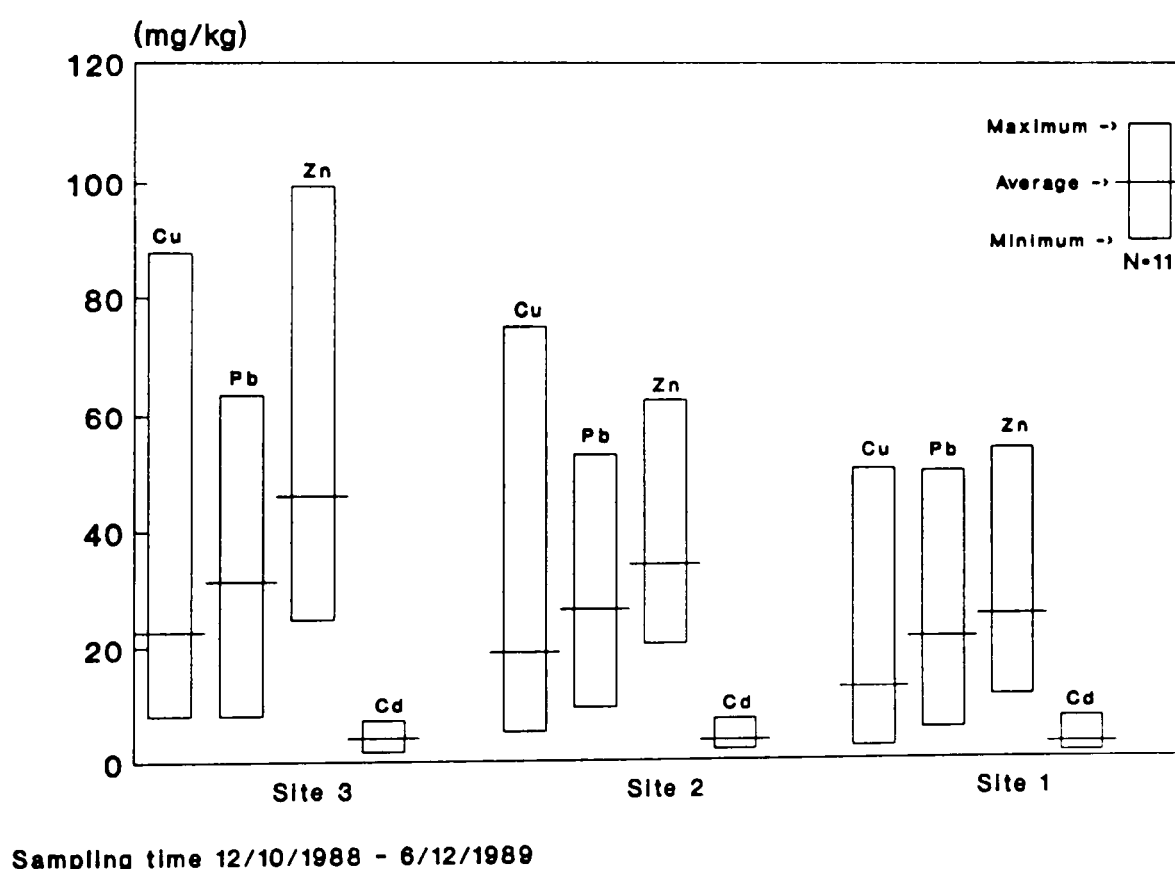


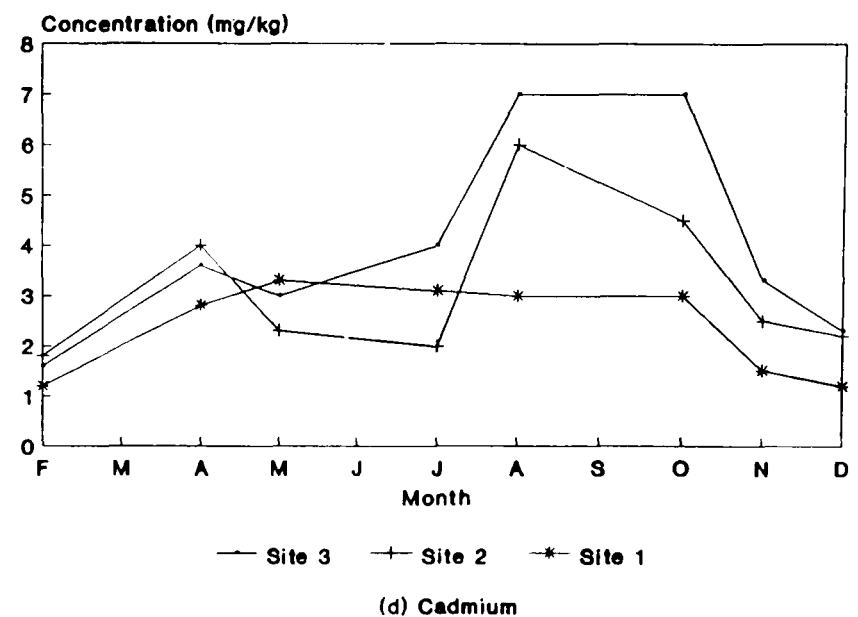
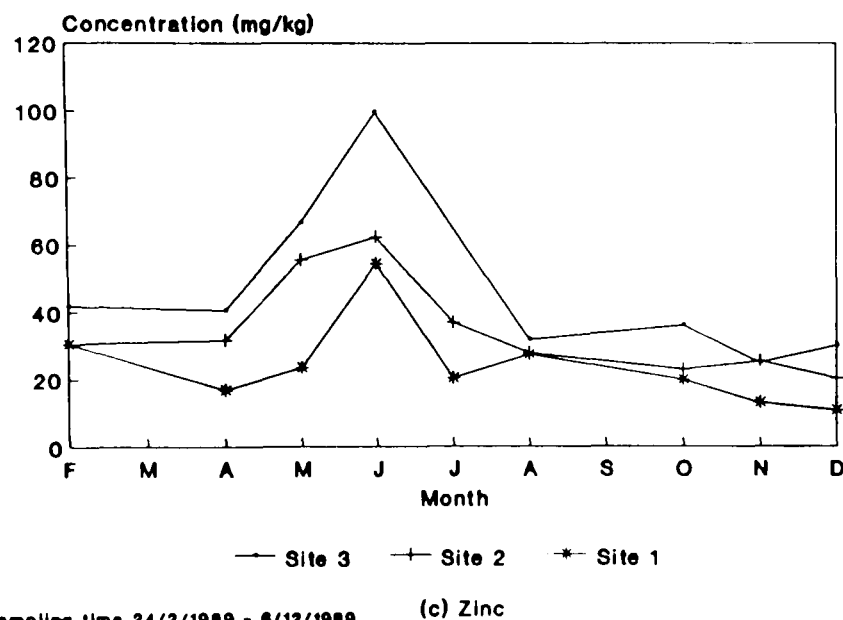
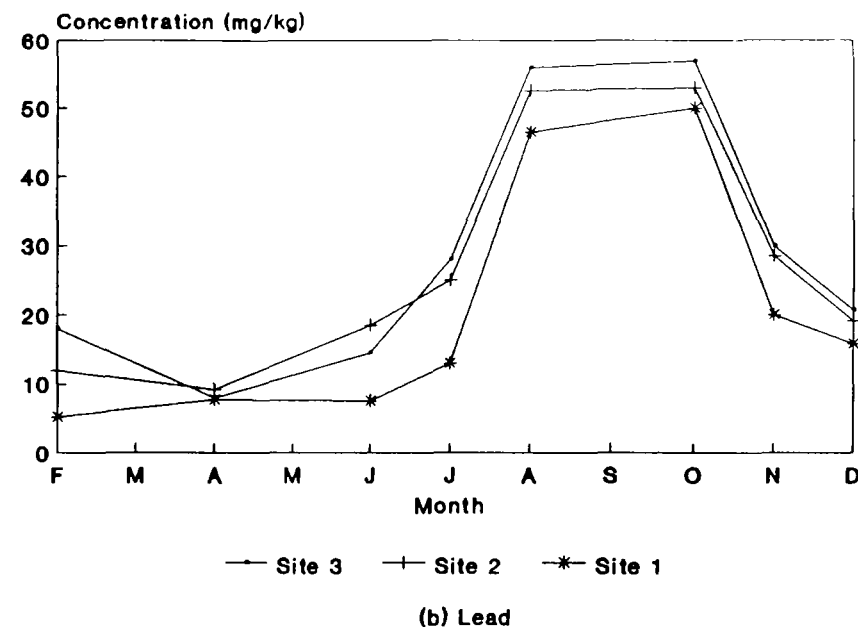
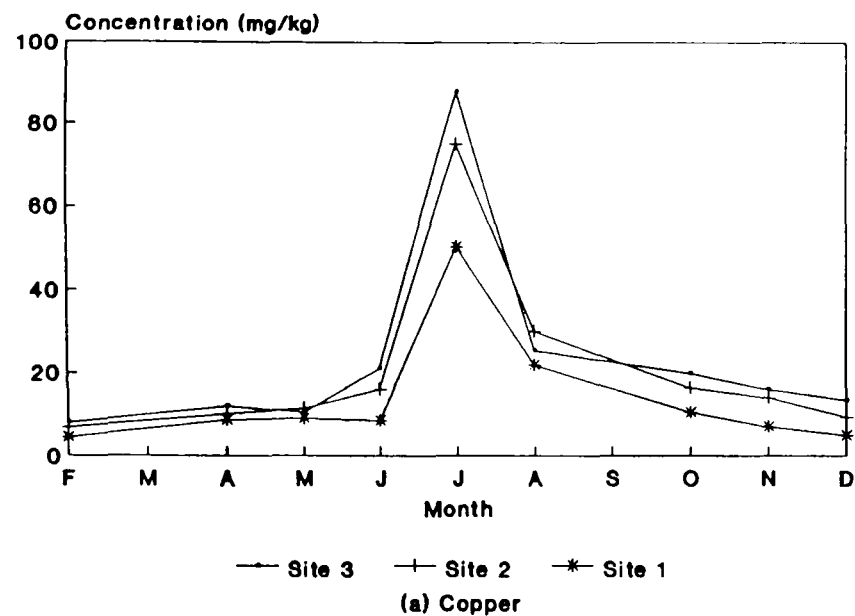
Fig.5.7 Average and ranges of heavy metal concentration in plant rhizome tissues

Table 5.7: T values of Typha rhizome samples
using Students 't' test

	$t_{0.05,11}$ (site 1 and site 2)	$t_{0.05,11}$ (site 1 and site 3)
Cu	3.326 (>1.796)	3.180 (>1.796)
Pb	2.852 (>1.796)	3.427 (>1.796)
Zn	2.805 (>1.796)	4.214 (>1.796)
Cd	2.401 (>1.796)	2.695 (>1.796)

when $n=11$, $P<0.05$, $t_{0.05,11}>1.796$, the difference is significant

All metals show temporal variability (Table 5.8) in Typha rhizome tissues with peak levels of Cu and Zn occurring during the growth period (Zn in June; Cu in July) and thus with an increase in demand for these associated plant micronutrients (Fig.5.8 a,c). There is general bioaccumulation of Cd and Pb in the rhizome during July to October (Fig.5.8 b,d). Rhizome Cu and Zn levels at the two urban sites are very similar to those obtained by Taylor and Crowder (1983) in Typha latifolia and Aulio (1980) in Nuphar lutea near a metal processing plant.



Sampling time 24/2/1989 - 6/12/1989

Fig.5.8 Temporal variations of heavy metal levels
in Typha rhizome tissues

Table 5.8: Average and standard deviation
of metal levels in Typha rhizome samples (mg/kg)

	Site 1	Site 2	Site 3
Cu	12.6 ± 12.9	19.0 ± 18.8	22.7 ± 21.2
Pb	21.5 ± 14.7	26.6 ± 13.7	31.6 ± 18.2
Zn	25.4 ± 12.1	34.3 ± 12.5	46.1 ± 21.2
Cd	2.6 ± 1.6	3.3 ± 1.7	3.9 ± 2.0

Plant rhizomes also participate in metal uptake from sediment although emergent species cannot significantly absorb metals directly through their thick rhizomes (Haslam, 1987). However, rhizomes play an important part in metal storage (see Chapter 6).

5.2.5 Heavy Metal Levels in Typha Leaf Tissues

Average metal concentrations together with ranges monitored during the 14 month field study for the leaf samples are shown in Fig.5.9. Although the metal leaf levels at the two urbanised sites (site 2 and 3) are higher than those of the rural site (site 1), the site 3 to site 1 mean concentration ratios decrease in comparison to root and rhizome. The ratio of metal levels in Typha leaf for the sites (3:2:1) are: Cu, 1.5:1.1:1, Pb, 1.1:1.2:1, Zn, 1.6:1.1:1 and Cd, 1.4:1.4:1. The differences in all metal levels between site 1 and site 3 are statistically significant. The differences in Pb and Cd levels between site 1 and site 2 are significant whereas Cu and Zn levels are not (Table 5.9).

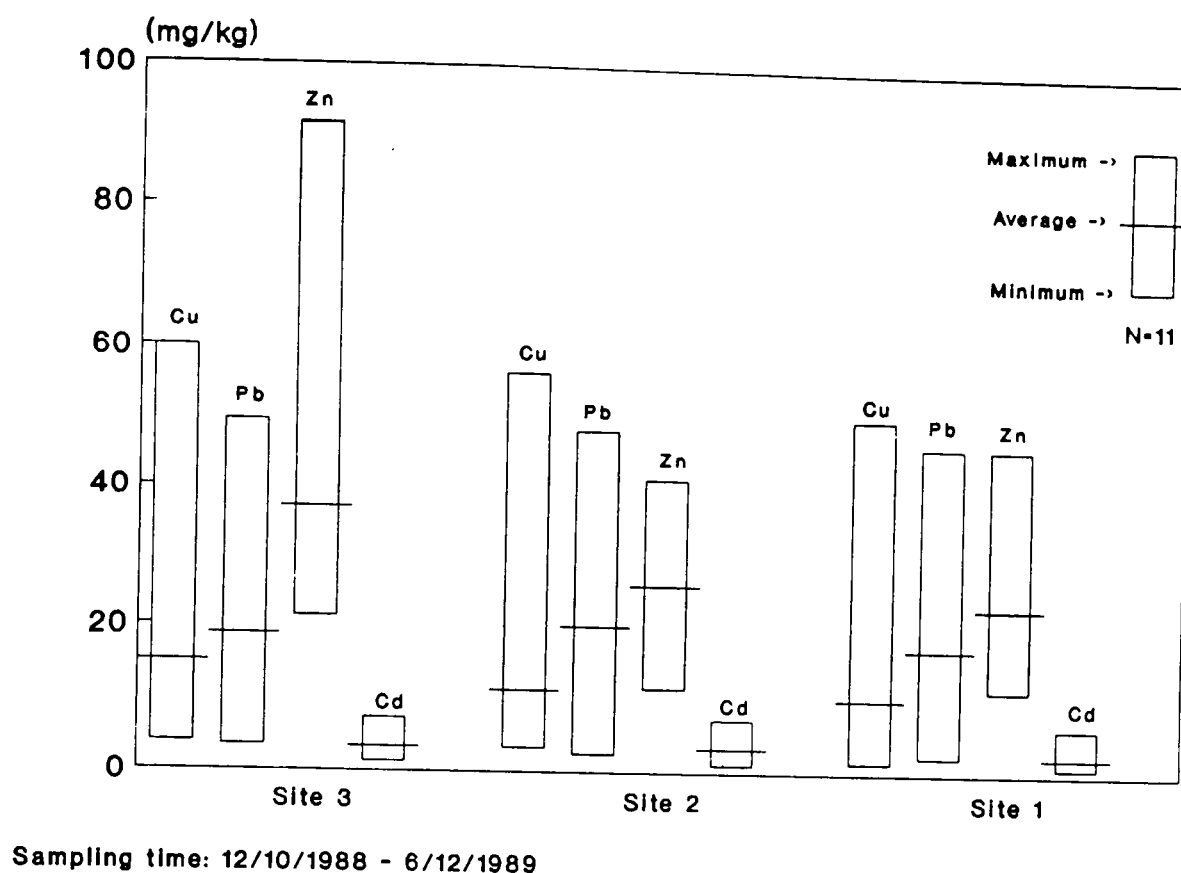


Fig.5.9 Average and ranges of heavy metal concentration in *Typha* leaf tissues

Table 5.9: T values of *Typha* leaf samples using Students 't' test

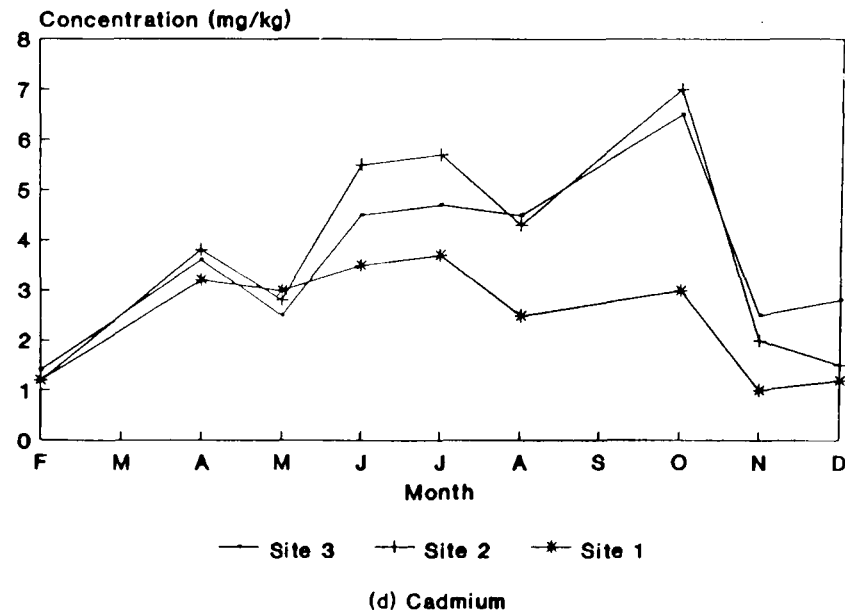
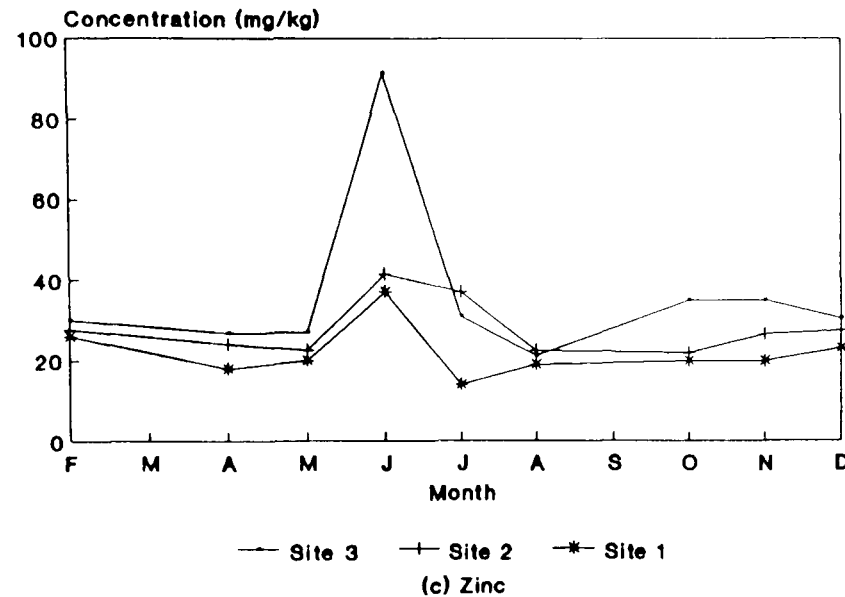
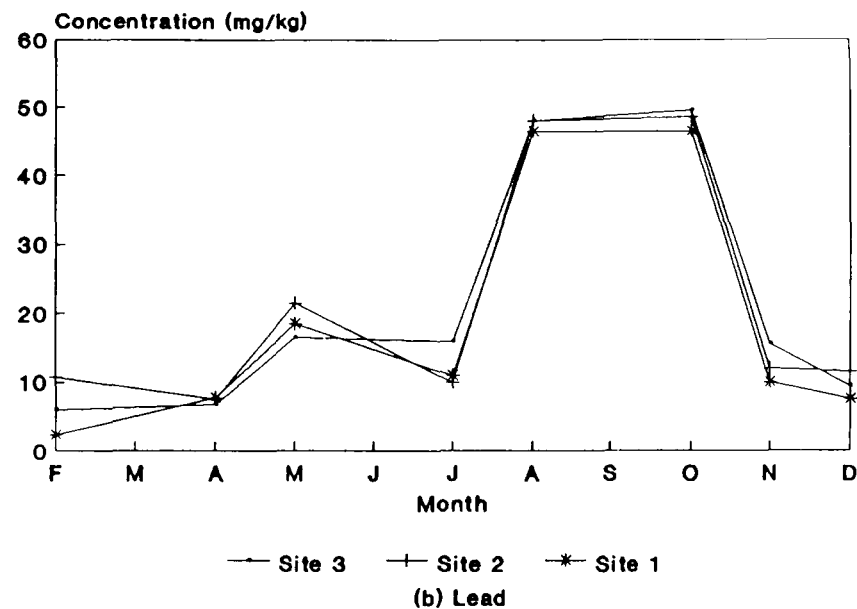
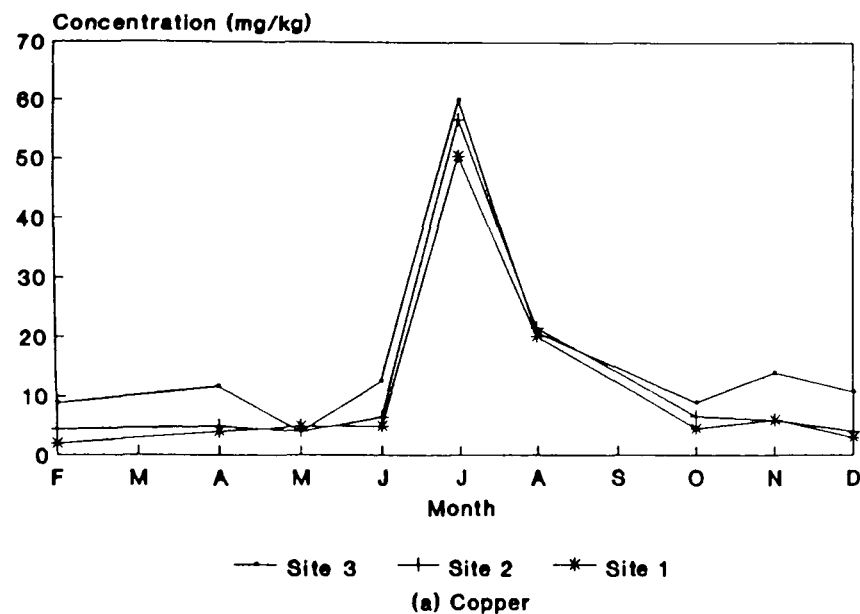
	$t_{0.05,11}$ (site 1 and site 2)	$t_{0.05,11}$ (site 1 and site 3)
Cu	1.591 (<1.796)	3.993 (>1.796)
Pb	2.248 (>1.796)	2.112 (>1.796)
Zn	0.685 (<1.796)	2.654 (>1.796)
Cd	2.640 (>1.796)	2.888 (>1.796)

when $n=11$, $P<0.05$, $t_{0.05,11}>1.796$, the difference is significant

All metals show temporal variability (Table 5.10) in Typha leaf tissues, with peak levels of Cu and Zn occurring during the growth period when both root and rhizome demand for these associated plant micronutrients is highest (Fig. 5.10 a,c). There is a progressive bioaccumulation of Cd in leaf tissue during May to October with peak levels being achieved in October (Fig. 5.10 d). Maximum leaf levels of Pb at all sites occur during August to October possibly due to translocation from the root tissues being augmented by atmospheric deposition as well as by increases in the permeability of the leaf during decomposition (Fig. 5.10 b). This latter mechanism has been suggested by Larsen and Schierup (1981) to work effectively for Phragmites which also showed temporal variability of Zn, Cu and Pb in leaf tissue.

Table 5.10: Average and standard deviation
of metal levels in Typha leaf samples (mg/kg)

	Site 1	Site 2	Site 3
Cu	10.4 ± 13.7	11.5 ± 15.1	15.1 ± 14.8
Pb	17.2 ± 15.0	20.1 ± 15.9	18.8 ± 15.3
Zn	23.2 ± 9.7	26.1 ± 7.5	37.0 ± 18.7
Cd	2.4 ± 1.6	3.4 ± 2.0	3.4 ± 1.9



Sampling time 24/2/1989 - 6/12/1989

Fig.5.10 Temporal variations of heavy metal levels
in Typha leaf tissues

The maximum leaf Pb levels (site 3) of 46.5 - 49.5 mg/kg are similar to those recorded by Reimer (1989) in a study which also showed considerable inter specific variation in macrophyte leaf Pb uptake. The average Cu levels of 15.1 and 11.5 mg/kg found at the two urban sites are similar to those recorded by Taylor and Crowder (1983). Zn levels 37.1 and 26.1 mg/kg at the two urban sites are higher than Taylor's results (18 ± 1 mg/kg) for a field study of metal uptake by Typha latifolia located near smelters. The average Cu level 10.4 mg/kg of site 1 are in the range of previous studies (of uncontaminated sites) on Typha latifolia and Typha angustifolia by Boyd (34 mg/kg, 1970 a), Mudroch and Capobianco (1.3 mg/kg, 1978), Mudroch (3-4 mg/kg, 1980), Reimer and Toth (6.3 mg/kg and 5.0 mg/kg, 1968), Varenko and Chuiko (24.3 mg/kg, 1971) and Guilizzoni (4.8 mg/kg, 1975).

Emergent species absorb little minerals through their aerial (leaf) parts with the main metal load found in plant leaf tissues arising from root uptake. The leaf metal yields have important implications for pollution control in terms of harvesting of the above-water foliage and this will be discussed later.

5.2.6 Comparison of metal levels in water, sediment and plant tissues

The concentrations of all metals in sediment and Typha are considerably higher than in the surrounding water. The ratios of sediment and Typha metal levels to surrounding water levels at site 3 are given in Table 5.11. The metal level ratios of Typha tissues to water range from 283 to 2389 whereas metal level ratios of sediment to water are

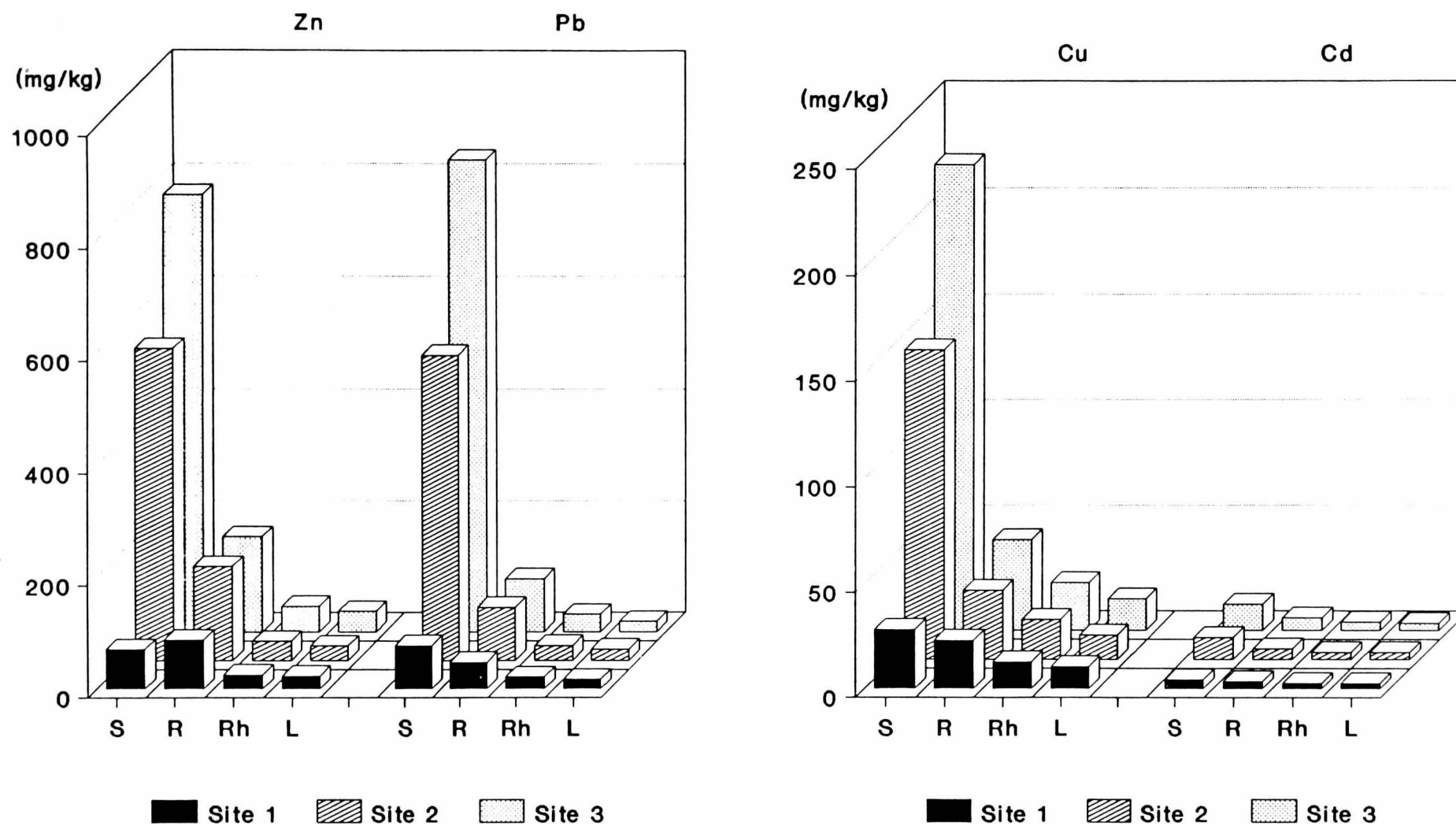
between 1393 and 21136. These results are consistent with those suggested by Merchyulenene and Nyanishkene (1976) and Leland and McNurney (1984).

Table 5.11: Metal level ratios of sediment and plant to water (site 3)

	Water (ug/l)	Root	Rhizome	Leaf	Sediment
Cd	1 (8.9)	674	438	382	1393
Cu	1 (53.4)	805	425	283	4137
Pb	1 (39.8)	2389	794	472	21136
Zn	1 (107.3)	1589	430	346	7259

The average metal concentrations monitored during the 14 month field study of sediment, root, rhizome and leaf samples at the three sites are shown in Fig.5.11.

There is a progressive decrease in Cu, Pb, Zn and Cd concentrations from sediment to Typha root, to rhizome and to leaf at all sites (except Zn at site 1). This is consistent with Taylor and Crowder's studies of Cu and Zn levels in Typha latifolia (except for Zn levels in leaf, 1983). The ratios of sediment to root to rhizome to leaf concentrations are given in Table 5.12. Welsh and Denny (1980) and Heisey and Damman (1982) have reported Pb root to leaf ratios of between 3.8 - 5.8 and 3 respectively for submerged macrophytes. In the present study root to leaf ratios are 2.7, 4.7 and 5.1 for site 1, 2 and 3 respectively.



S:sediment, R:root, Rh:rhizome, L:leaf
Sampling time 12/10/1988 - 6/12/1989

Fig.5.11 Heavy metal levels in sediment and Typha tissues

Table 5.12: Sediment to leaf metal average concentration ratio

		sediment	:	root	:	rhizome	:	leaf
		(mg/kg)						
Site 3	Cu	14.6		2.8		1.5		1 (15.1)
	Pb	44.8		5.1		1.7		1 (18.8)
	Zn	21.0		4.6		1.2		1 (37.1)
	Cd	3.6		1.8		1.1		1 (3.4)
Site 2	Cu	12.8		2.9		1.7		1 (11.5)
	Pb	27.1		4.7		1.3		1 (20.1)
	Zn	21.3		6.4		1.3		1 (26.1)
	Cd	3.5		1.7		1.1		1 (3.4)
Site 1	Cu	2.7		2.2		1.2		1 (10.4)
	Pb	4.5		2.7		1.3		1 (17.2)
	Zn	3.0		3.8		1.1		1 (23.2)
	Cd	1.7		1.4		1.1		1 (2.4)

Although root to leaf mean concentration ratios increase for Cu, Pb, Zn and Cd at the urban sites, the ratios of rhizome to leaf concentrations are relatively constant for all metals at the three sites which range between 1.1:1 for Cd to 1.7:1 for Pb and Cu. This suggests that the ultimate tissue distribution is probably constitutionally determined and is independent of root metal uptake rates. Taylor and Crowder (1983) have shown that Zn and Cu concentrations of Typha leaves were not correlated with sediment concentrations.

Sediment to leaf mean Pb concentration ratios increase in the order of 4.5:1, 27:1 and 44.8:1 at sites 1, 2 and 3 respectively, indicating the affinity of Pb for the

sediment particulate phase. However, a similar trend is observed for Zn, which is known to occur predominantly in the more bioavailable dissolved phase. The sediment to leaf mean metal concentration ratios are in the order of Pb>Zn>Cu>Cd at all sites. This implies that the uptake ability of Typha plants is in the reverse order of Cd>Cu>Zn>Pb.

5.3 Conclusions

The major findings of this section of the research can be summarised as follows:-

a) An increase in sediment and Typha tissue levels is observed from the rural to urban sites with site 3 to site 1 sediment average concentration ratios ranging from 3:1 for Cd to 11:1 for Zn. The range of site 3 to site 1 root average concentration ratios varies from 1.8:1 for Cd to 2:1 for Pb.

b) The statistical analysis shows that the water concentration differences of all metals between site 3 and site 1 are significant. The concentration differences of Cu and Cd between site 2 and site 1 are significant whereas the differences of Pb and Zn are not. The metal concentration differences in sediment, Typha root and rhizome between urban sites and the rural site are also significant as are the leaf concentration differences of all metal levels between site 1 and site 3. The differences of Pb and Cd levels between site 1 and site 2 are significant whereas Cu and Zn levels are not.

c) Field samples from all sites show a progressive decrease in metal levels from sediment to root to rhizome

to leaf, with sediment to leaf ratios increasing in the case of Pb from 4.5:1 to 44.8:1 from site 1 to 3. Although root to leaf mean concentration ratios increase for Cu, Pb, Zn and Cd at the urban site, the ratio of rhizome to leaf concentration is relatively constant for all metals at all sites. This suggests that the ultimate tissue distribution is probably metabolically determined and is independent of root metal uptake rates. The sediment to leaf mean metal concentration ratios are in the order of Pb>Zn>Cu>Cd at all sites. This implies an uptake ability of Typha plants in the reverse order of Cd>Cu>Zn>Pb.

d) All metals show temporal variability in Typha tissues, with peak levels of Cu and Zn in root, rhizome and leaf coinciding with the growth period and thus with an increase in demand for these associated plant micronutrients. There is bioaccumulation of Cd and Pb during August and October in root, rhizome and leaf tissues. The temporal variability in accumulation rates, especially for leaf tissue, has important management implications in terms of heavy metal removal through by harvesting the standing leaf crop.

Chapter 6: The Comparison of Field Studies and Greenhouse Dosing Experiments and their Management Implications

6.1 Introduction

Emergent aquatic macrophytes can take up heavy metals from sediments via root systems and translocate them to the rhizome and aerial parts. However, the greater part of heavy metals taken up by the plants is retained in the roots and rhizome with smaller amounts being translocated to the above - ground sections of the plant (Larsen and Schierup, 1981). Heavy metals stored in the above - ground part can be removed by cropping the leaves. Black et al. (1979, 1984) reported that cutting and harvesting Typha latifolia leaves can remove 25 to 42 % of the Zn accumulated in the plants and prevents the leaching of the metal from associated decaying organic matter.

The aim of this study was to compare metal loads found in Typha latifolia resulting from both greenhouse-based dosing experiments and from field studies. Heavy metal distributions found in Typha have already been analysed and the management implications of heavy metal removal by cropping plant leaves are discussed further in this section.

6.2 The comparison of field studies and greenhouse experiments

6.2.1 The comparison of sediment metal levels

The average metal levels in the sediments at three field sites and metal levels in the dosed peat from greenhouse

experiment after 8 weeks have been compared with the unpolluted and toxic levels found in soil which were given in Table 2.2 (Romero et al., 1989 and Baker and Brooks, 1989). An evaluation of toxic metal levels in sediment can be derived from inspection of Table 6.1. All metal levels, with the possible exception of Cd, found at site 1 are well below toxic levels with Cu and Zn levels being very close to the unpolluted metal levels of background soils. Sediment Cu, Pb, Zn and Cd levels at both urban sites are very much higher than the generally quoted toxic levels with the highest ratio (metal levels of urbanised sites investigated to general toxic levels in soil) being for Pb (5.5 and 8.4 at site 2 and 3, respectively). This reflects the generally more contaminated and toxic nature often associated with urban discharges (Ellis, 1986). The relative toxicities of the metals are in the order of Pb>Cd>Zn>Cu for site 2, and Pb>Cd>Cu>Zn for site 3.

Table 6.1: The toxicity of metal levels in sediment

Soil (mg/kg)			Sediment investigated (mg/kg)			
Unpolluted*	Toxic*		Site 1	Site 2	Site 3	Dosed Peat (Surface)
levels	levels					
				Ratio ⁺	Ratio ⁺	Ratio ⁺
Cu	25	100	28	147(1.5)	220(2.2)	187(1.9)
Pb	25	100	78	545(5.5)	841(8.4)	168(1.7)
Zn	65	300	71	557(1.8)	779(1.6)	295(1.0)
Cd	1	5	4	11(2.2)	12(2.4)	286(57.2)

* from Table 2.2

+ ratios of site 2 : toxic, site 3 : toxic and dosed : toxic

Copper, Pb and Cd levels in dosed peat after the 8 weeks dosing experiment are all higher than threshold toxic levels especially for Cd. Pb levels are much lower than the levels found at the two urban sites. The Cu is similar to the levels found in urban sites whereas Cd level is much higher than the urban sites. Zn levels in dosed peat are similar to the toxic level and fall between the levels of the rural and urban sites. The relative toxicity of the metals in dosed peat is in the order of $Cd > Cu > Pb > Zn$.

The maximum metal loads found in the sediments of the greenhouse experiments and field studies are given in Table 6.2. Copper, Pb and Zn levels for the two urban sites sediments are very much higher than the peat metal levels obtained following the 8 week dosing experiments (Cu levels at site 2 are similar to the results obtained for the dosing experiments). Cd levels however are much lower than the levels obtained for the dosed peat. This suggests that the input rates of metal loads received at the urban sites are high and exceed the potential rate of uptake and removal by Typha and therefore the metals accumulate in the sediment phase.

Table 6.2: Maximum Metal Loads in Sediment
for Dosing Experiments and Field Studies

	Dosed (surface)	Site 3	Site 2	Site 1
Cu	187	304	178	42
Pb	168	1199	784	97
Zn	295	978	764	123
Cd	286	30	15	13

* values would be in mg if based on extraction from 1kg dry weight
ie mg/kg equivalents

6.2.2 The comparison of Typha tissue metal levels

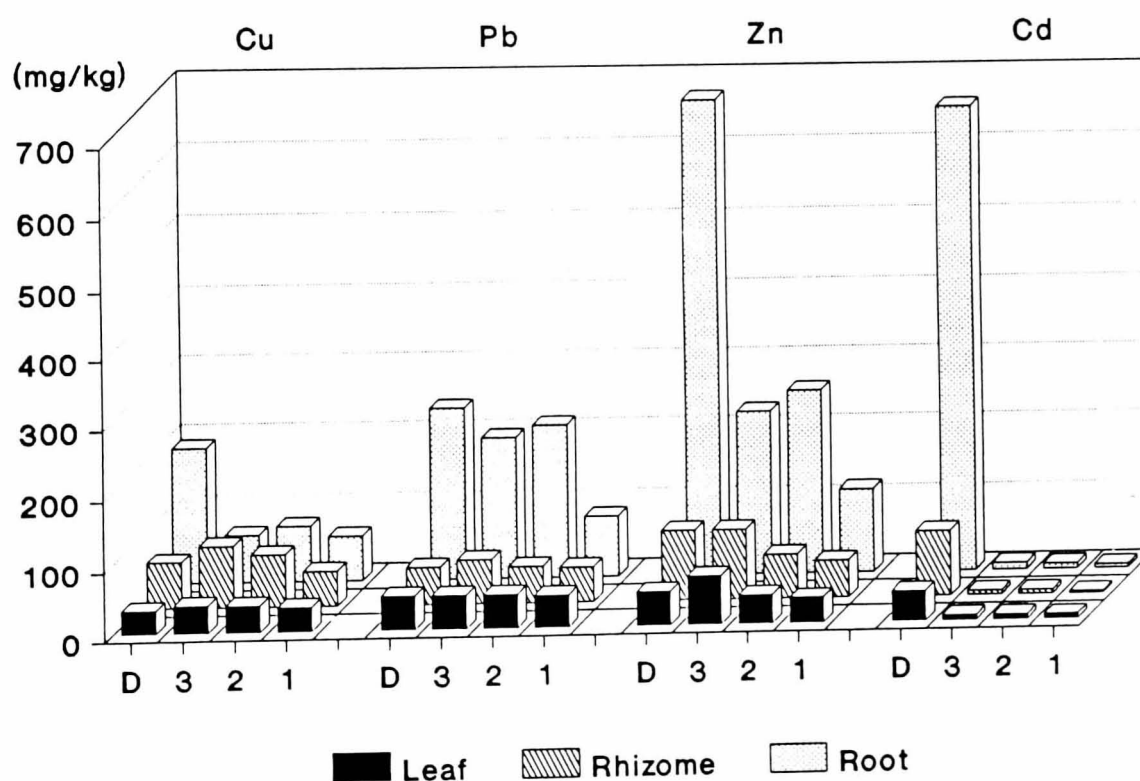
The average metal levels in plant tissues at three field sites and from greenhouse dosing experiments have been compared with the general unpolluted and toxic levels found in plants which were quoted in Table 2.2 (Table 6.3). Only Cu and Cd levels at site 3 reach the general toxic levels whilst all others fall below the indicated toxic levels. This is probably due to the avoidance or restriction of metal uptake in tolerant races of plant (Baker and Walker, 1990). The rhizome and leaf metal levels for field sites are all below the general toxic levels. Metal levels in dosed plant root are however much higher than the general toxic levels in the plant; Cu and Cd levels in Typha rhizome and Cd levels in the leaf also exceeded the toxic level. This demonstrates the high tolerance of metal toxicity for these species.

Table 6.3: The toxicity of metal levels in plant

Plant (mg/kg)	Cu	Pb	Zn	Cd
Unpolluted*	12	10	60	
Toxic levels*	40	150	400	5
Root				
Site 1	23.0	47.3	87.5	3.3
Site 2	32.7	94.9	160.1	5.0
Site 3	39.7	85.6	153.5	5.4
Dosed	190.0	242.0	689.0	662.5
Rhizome				
Site 1	12.6	21.5	25.4	2.6
Site 2	19.0	26.6	34.3	3.3
Site 3	22.7	31.6	46.1	3.9
Dosed	66.0	54.2	99.5	92.7
Leaf				
Site 1	10.4	17.2	23.2	2.4
Site 2	11.5	20.1	26.1	3.4
Site 3	15.1	18.8	37.0	3.4
Dosed	23.5	38.0	36.6	43.8

* from Romero et al., 1989, Baker and Brooks, 1989

The maximum metal loads in Typha root, rhizome and leaf for the greenhouse experiments and field studies are given in Fig. 6.1. The maximum root Pb levels at the two urban sites are similar to the root Pb levels obtained after the 8 weeks dosing experiment, whereas Cd, Cu and Zn levels are much lower than those of the dosed root. The maximum Cu, Pb and Zn levels in Typha rhizome and leaf levels of the two urban sites are similar to those obtained from the greenhouse dosing experiments, whereas the Cd levels are much lower.



D:Dosed, 3:Site 3, 2:Site 2, 1:Site 1

Fig.6.1 The comparison of Typha latifolia tissue metal levels in dosing experiments and field studies

The comparison of maximum Typha tissue metal levels for the field studies with the long term dosing experiments suggests that the limit of uptake for Cu, Pb and Zn is probably achieved at the urban sites (except Cu and Zn levels in root tissue) but has not yet been achieved in the case of Cd.

6.2.3 Metal uptake efficiency of Typha latifolia

Both short term and long term greenhouse-based metal uptake experiments with Typha latifolia show that the efficiency of Typha root metal uptake is in the order of $Cd > Cu > Zn > Pb$. The field studies of Typha metal uptake show the same results. These results are in agreement with those obtained by Harrison and Chirgawi (1989). In his study of soil and air as contributors of trace metal to terrestrial plants, it was suggested that the efficiency of uptake of metals from soil was generally high for Zn and Cd but low for Pb.

6.3 Management implications for heavy metal removal

6.3.1 Biomass measurement

The biomass of root, rhizome and leaf of Typha from site 1 during October, 1989 were measured and the results are shown in Table 6.4 below. The ratios of tissue biomass show that Typha root tissue biomass is relatively low in comparison to rhizome and leaf; hence its negligible role as a metal storage tissue. The highest biomass was found in Typha rhizome tissue. The rhizomes play an important role in metal storage (see section 6.3.2) with 42.4% of Typha plant biomass coming from leaves. This has important implications for metal removal by cropping (see section

6.3.3). The calculated leaf biomass (8381 kg/ha) and plant total biomass (19,761 kg/ha) are similar to those obtained by Blake et al. (9150 and 16,169 kg/ha for leaf and plant total biomass respectively), for their study of Typha latifolia on sediment contaminated with Zn (1987).

Table 6.4: Dry Weight Tissue Biomass (October, 1989, Site 1)

Biomass	kg/ha	Ratio	% of total
Root	604	1	3
Rhizome	10,776	18	54.5
Leaf	8381	14	42.5
Total	19,761	33	100

The Typha leaf biomass (site 1) during the growing season was also measured and the results are shown in Table 6.5. The leaf tissue biomass increases from May to October with the highest leaf tissue biomass being found in July.

Table 6.5: Dry Weight Leaf Tissue Biomass (1989, Site 1)

	May	June	July	October
Leaf (kg/ha)	2400	7433	8740	8381

6.3.2 Metal distribution in plant tissues

When metal levels are converted to tissue loadings using the dry weight biomass tissue ratios for Typha collected from the field sites (root:rhizome:leaf=1:18:14, Table 6.4), the resultant metal distributions are as shown in Fig.6.2.

The metal loading data clearly indicate that the major metal bioaccumulation target area in Typha is the rhizome with 54-61 % metal load being stored in this area. Approximately one-third of the metal load is stored in Typha leaf whereas only 4-12 % is stored in Typha roots. These results show that the major burden of heavy metal taken up by Typha is retained in roots and rhizomes (60, 67, 68 and 66 % for Cd, Cu, Pb and Zn respectively). This is in broad agreement with Blake et al.'s (1987) study which showed 58-75 % Zn of accumulated by Typha being stored in roots and rhizomes. However, Larsen and Schierup's study (1981) with Phragmites showed a much higher metal storage in roots and rhizomes (77-97 %). Typha accumulates some 32-40 % of metals in leaf tissue (Fig.6.2) and these metals could potentially be released by the decomposition processes (Black, 1979, 1984). It might therefore be necessary to cut and harvest the plant, as this technique would allow the export of 30-40 % of the metals accumulated in Typha latifolia and prevent the leaching from decaying organic matter of the metal back into the water phases.

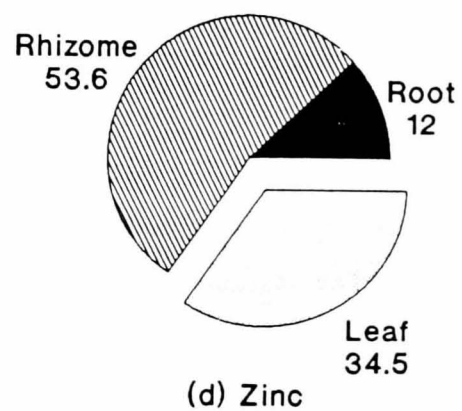
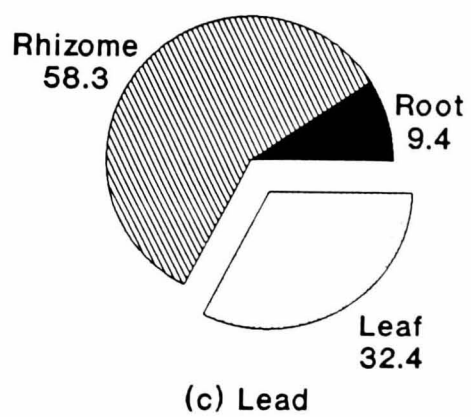
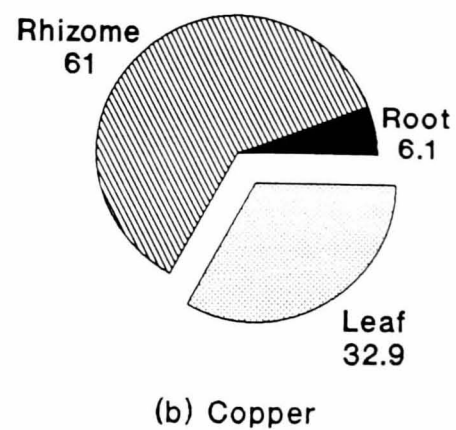
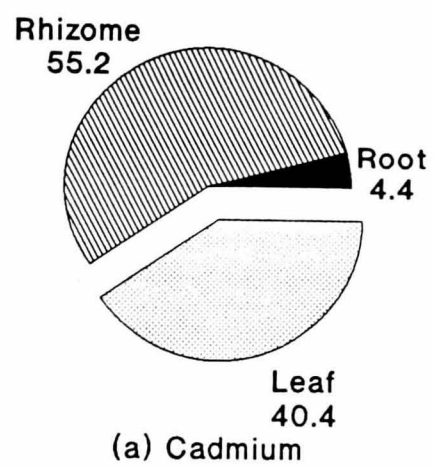


Fig.6.2 Metal load distributions in Typha tissue (%)

6.3.3 Heavy metal removal

The partial removal of heavy metal pollutants in macrophytes can be achieved by harvesting of the foliage where the accumulated metals are stored. The variation the optimal cropping months for each metal, assuming one crop per year, is given in Table 6.6.

These results have some important management implications. An annual crop of emergent leaf biomass, preferably undertaken between June to October, would provide an efficient method of heavy metal removal from recently constructed wetlands. In mature wetlands however, preferential metal accumulation in the sediment store will inevitably reduce the value of metal removed by leaf harvesting. This is due to the unavoidable disturbance of the benthal substrates and consequent release of metals which will subsequently effect the potential advantages that might accrue from metal removal by cropping. Furthermore, any disruption of the submerged leaf tissue matrix would interfere with the absorption of metals from the water phase and their transfer to the rhizome, which Mortimer (1985) considers to be an important pathway.

Table 6.6: Maximum Dry Weight Leaf Metal Loads (g/ha)

	Cd	Cu	Pb	Zn
Optimal Cropping Month	October	July	October	June
Site 1	25.1	440.5	389.7	275.0
Site 2	58.7	494.7	406.5	308.5
Site 3	54.5	524.4	414.9	680.0

6.4 Conclusions

The major findings of this section of the research can be summarised as follows:-

a) The field study of sediment and plant samples indicates that all metal sediment levels at the urbanised sites are higher than the generally recorded toxic level. The toxic levels are found in the order of Pb>Cd>Zn>Cu for site 2, and Pb>Cd>Cu>Zn for site 3. These results indicate that Typha latifolia is a metal tolerant species and can thrive on metal contaminated sediment in urban wetlands. All sediment metal levels at the background control site 1 are below general background toxic levels with Cu and Zn levels being very close to the unpolluted metal levels found in soil.

b) Copper and Cd root levels at site 3, as well as Cd root levels at site 2 reached metal levels in Typha tissues which are regarded as potentially toxic to plants. Metal levels in dosed plant root are all much higher than the general toxic levels; Cu and Cd levels in Typha rhizome and Cd level in leaf also exceed the toxic level. This indicates that Typha latifolia cannot only survive in metal contaminated sediment but also can efficiently and rapidly uptake metals especially Cu and Cd to high levels.

c) The comparison of greenhouse experiments and field studies shows that under field conditions, heavy metals can accumulate in sediment to high levels. This result demonstrates the important role of the sediment store as a metal sink in polluted receiving waters.

d) This study also shows that the limit of Typha latifolia metal uptake for Cu, Pb and Zn is probably achieved at urban sites over fairly short time scales, but it is rather more difficult to achieve saturation in terms of Cd uptake. The metal uptake efficiency of Typha roots for both greenhouse experiments and field studies show the order to be Cd>Cu>Zn>Pb.

e) The biomass measurement and plant metal distribution analysis show that Typha rhizome is the prime target area for metal storage (54 - 61 %). Plant leaves can store up to 32 - 40 % of metals accumulated in the plant tissue. These results can have important management implications.

f) Heavy metal removal can be achieved by cropping plant leaves. An annual crop of Typha leaf biomass, preferably undertaken in June to October can remove up to Cu 524, Pb 415, Zn 680 and Cd 59 g/ha. This could provide a useful method of heavy metal removal from recently constructed wetlands.

Chapter 7: Conclusions

7.1 Summary of major findings

a) The research shows an efficient metal uptake (Cu 25.1, Pb 20.3, Zn 32.5 and Cd 31.5 mg/kg/wk) by substratum(peat) in greenhouse-based phytoassays which is confirmed by the very high metal levels determined in urban field sediment studies. Levels found in urban sediment are 10^3 - 10^4 times higher than the metal levels encountered in the water column and 1.5 - 8.4 times higher than the reported sediment toxic levels. This indicates that sediment plays a very important role as both a metal sink and reservoir for leaf decomposition as well as for contaminants contained in urban surface runoff discharges.

b) The research demonstrates effective heavy metal Cu, Pb, Zn and Cd uptake by Typha latifolia. The uptake efficiency of Typha roots for both greenhouse experiments and field studies were found to be in the order of Cd>Cu>Zn>Pb. The study also indicates that the limit of Typha latifolia metal uptake as determined by dosing experiment for Cu, Pb and Zn is probably achieved at the urban sites (except Cu and Zn levels in root tissue), but is not yet reached for Cd.

c) Both greenhouse experiments and field studies show that metal levels in Typha latifolia can reach levels which exceed the suggested toxic metal levels of plant tissues. The tolerance of metals is in the order of Cd>Cu>Pb>Zn. This indicates that Typha latifolia plants not only can survive in a highly metal contaminated sediment matrix but can also accumulate heavy metals especially Cd and Cu to high levels in the plant tissues. These results

demonstrate the high tolerance these species have for the metal toxic levels normally encountered in the environmental ecosystem.

d) All metals show marked temporal variability in Typha tissues, with peak levels of Cu and Zn found during the growth period when there is an increase in demand for these associated plant micronutrients. There is bioaccumulation of Cd and Pb during August and October in plant tissues. This temporal variability in metal levels especially for leaf components has important management implications by means of cropping plant leaves for heavy metal removal from urban wetlands.

e) The comparison of dosing experiments of Typha latifolia and Juncus effusus shows that Typha roots have a higher potential for metal uptake. The results also show that the metal translocation from root to leaf is more efficient in Typha for Cd, Cu and Pb, whereas Juncus is more efficient for Zn translocation.

f) The biomass measurements and plant metal distribution analysis show that Typha rhizome is the major target area for metal storage. Typha rhizomes can store 54 - 61 % of the metals accumulated in the plant tissues; plant leaves store some 32 - 40 % metals.

g) Heavy metal removal from urban wetlands can be achieved by cropping plant leaves. An annual crop of Typha leaf biomass, preferably undertaken in June - October can remove up to Cu 525, Pb 415, Zn 680 and Cd 59 g/ha. This can provide a useful method of heavy metal removal especially in recently constructed artificial wetlands.

h) The results of this study provide useful information and methods for water quality improvement of urban runoff, which are especially valuable in terms of bioengineering design of urban flood storage ponds.

7.2 Suggestions for further work

a) The heavy metal concentrations presented in this study are of total metal concentrations. The sediment - metal speciation and their relationship with plant - metal uptake have not been studied. The study of such sediment - metal speciation would provide further useful information about bioavailable metals present in sediment and reveal more detailed relationships on plant - metal uptake.

b) Chapter 4 has outlined the results of greenhouse dosing experiments carried out with static tanks. Further studies should be taken to examine heavy metal uptake by sediment and plant under flow-through conditions in both the laboratory and field. This would model heavy metal uptake in urban aquatic ecosystems and their mechanisms under more realistic and mobile conditions.

c) The studies described in chapter 5 have indicated the metal uptake ability of Typha. In order to understand the heavy metal cycle in an aquatic environment, it is very important to know the plant decomposition process and heavy metal release. Further work should be undertaken to study the release mechanisms and controls of heavy metals from decomposing plant materials under both laboratory and field conditions.

d) The studies described in chapter 6 have indicated that the limit of Typha latifolia metal uptake examined by

dosing experiments for Cu, Pb and Zn is probably achieved at urban sites, but is not yet reached for Cd. Further work should be taken to study the factors which effect the plant metal uptake under both greenhouse dosing experiment and field conditions.

e) In order to examine the performance of macrophytes for metal uptake in urban wetlands and their role for water quality improvement in urban areas, controlled field site studies should be undertaken taken to determine the role of planted macrophytes species in both metal uptake and removal in artificially constructed urban wetlands. These studies could provide useful criteria for bioengineering design and urban wetlands management.

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This thesis is dedicated to my daughter Wang Xiaopei who is living with grandparents in China in the hope she will understand why she has missed her mother's love in the last two years and also in the hope she may grow up in a world which is environmentally friendly and full of freedom and happiness.

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APPENDIX A.

The following co-authored papers were published during the course of the research project:

A2 - A8 The Use of Macrophytes for Water Pollution Control in China and the UK. IAWPRC Specialist Group: The Use of Macrophytes in Water Pollution Control, Newsletter No.3, March 1990, pp 25 - 31

A8 - A17 Metal Uptake and Associated Pollution Control by Typha latifolia in Urban Wetlands. In: P.F. Cooper and B.C. Pindlater (Edits) Proceedings of International Conference on Use of Constructed Wetlands in Water Pollution Control, September 1990, pp 451 - 459